

Species diagnosis, hosts and distribution of cyst nematodes of the genus *Heterodera* (Tylenchida: Heteroderidae) with a focus on species of concern for Australia

Daniel C. Huston, Manda Khudhir, Sarah Dunstan, and Mike Hodda

Australian National Insect Collection, National Research Collections Australia, CSIRO, PO Box 1700, Canberra, ACT 2601, Australia

Daniel.Huston@csiro.au

Cyst nematodes of the genus *Heterodera* include around 80 species of plant-parasites, many of which are significant pests of important crops. Species of *Heterodera* are differentiated by subtle morphological differences making diagnoses difficult. We are studying the species richness, distribution, and host range of cyst nematodes in Australia, which includes putative endemics, potentially undescribed natives, and quarantine exotics, with an aim to improve diagnostic capacity for these pests in Australia and the world.

Evaluating publicly available barcoding data

Morphological taxonomy of cyst nematodes is complex
Diagnostics is therefore becoming increasingly reliant on molecular data

Around 40% of species have no molecular data available

Some of the more commonly used gene regions routinely fail to distinguish between some species

Presently available data used

All 18S, ITS, 28S and *cox1* sequences on NCBI GenBank for all species of *Heterodera* to August 2021.

Results

Rate of obviously erroneous sequences is fairly low

All four molecular markers failed to differentiate between at least one species pair

A combination of multiple markers is best for species identification

The *cox1* marker is most useful for species differentiation where resources are limited.

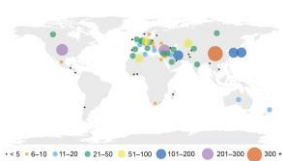


Figure 1: Geographic distribution of sequences evaluated in our study. Circles indicate number of sequences from a region.

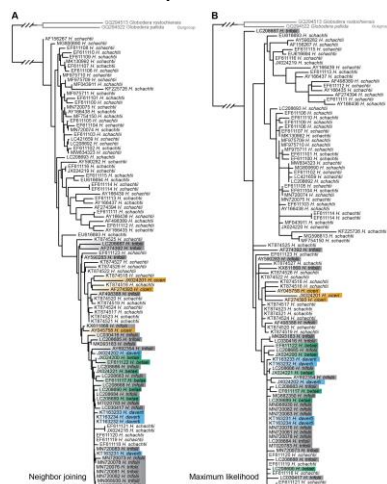


Figure 2: Neighbour joining (A), and Maximum likelihood (B) trees derived from analyses of ITS gene sequences for the Schachtii species group showing multiple instances where species cannot be differentiated.

Building a reference collection of *Heterodera* species for biosecurity diagnostics

Multiple *Heterodera* spp. are National Priority Plant Pests in Australia
Few specimens are held in Australian collections

We are building a new collection of cyst nematodes as a resource for biosecurity diagnostics and taxonomic research.

We seek contact and collaboration to obtain or exchange specimens of:

- many species and populations of *Heterodera* cyst nematodes; and
- from as wide a geographic distribution as possible.

We aim to build a collection with diagnostic utility, to tackle multiple taxonomic questions and issues of identity, expand DNA sequence coverage for the genus enhancing molecular diagnostic testing, and ultimately produce a National Diagnostic Protocol for the genus *Heterodera*.



Figure 3: Soil collections from over 100 sites across multiple provinces in Turkey, made by a team of our collaborators led by Prof. Ian T. Riley (Niğde Ömer Halisdemir Üniversitesi). This collection includes many species of *Heterodera*. Image courtesy of Prof. Ian T. Riley.



Figure 4: Cysts extracted from the soil in Figure 3 by our Turkish collaborators and subsequently transferred to the Nematology collection at the Australian National Insect Collection (CSIRO). From this collection we are generating molecular data and producing morphological specimens for the collection. Image courtesy of Prof. Ian T. Riley.

Distribution and species richness of *Heterodera* in Australia

Multiple species of *Heterodera* have been reported from Australia but knowledge of presently occurring species and their distributions are lacking.

We are making new field collections and reevaluating existing archived soil collections to determine the accuracy and validity of Australian records using an integrated morphological and molecular approach. We have detected several species not previously known from Australia, fortunately none of these are priority pest species.

Additional projects include:

- Determining if *Heterodera avenae* and *Heterodera australis* co-occur in Australia.
- Attempting to recollect *Heterodera graminis* from the type-locality in New South Wales, Australia.
- Working with the Australian hops industry to determine the distribution of *Heterodera humuli*.

Manuscripts reporting our findings are in the works – so watch this space!



Figure 5: 'cereal cyst nematode' damage to cereals in Western Australia. This nematode has long been assumed to be *Heterodera avenae*, however our preliminary molecular analyses suggest all populations are likely *Heterodera australis*. Image credit: Vivian Vanstone, Department of Agriculture and Food, Western Australia.

Seeking collaborators!

Many species of *Heterodera* will only be definitely distinguishable with more data on intraspecific and interspecific variation in morphological and molecular characters, so we are seeking collaborators for collecting specimens to facilitate acquisition of said data, and ultimately to better define what is likely to make a diagnosable species in the genus. If you think you may be able to share specimens with us, facilitate or make new collections, please get in contact with us. We have a budget to assist in these activities!

FOR FURTHER INFORMATION:

Dr Daniel C. Huston; Dr Mike Hodda
Daniel.Huston@csiro.au; Mike.Hodda@csiro.au

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