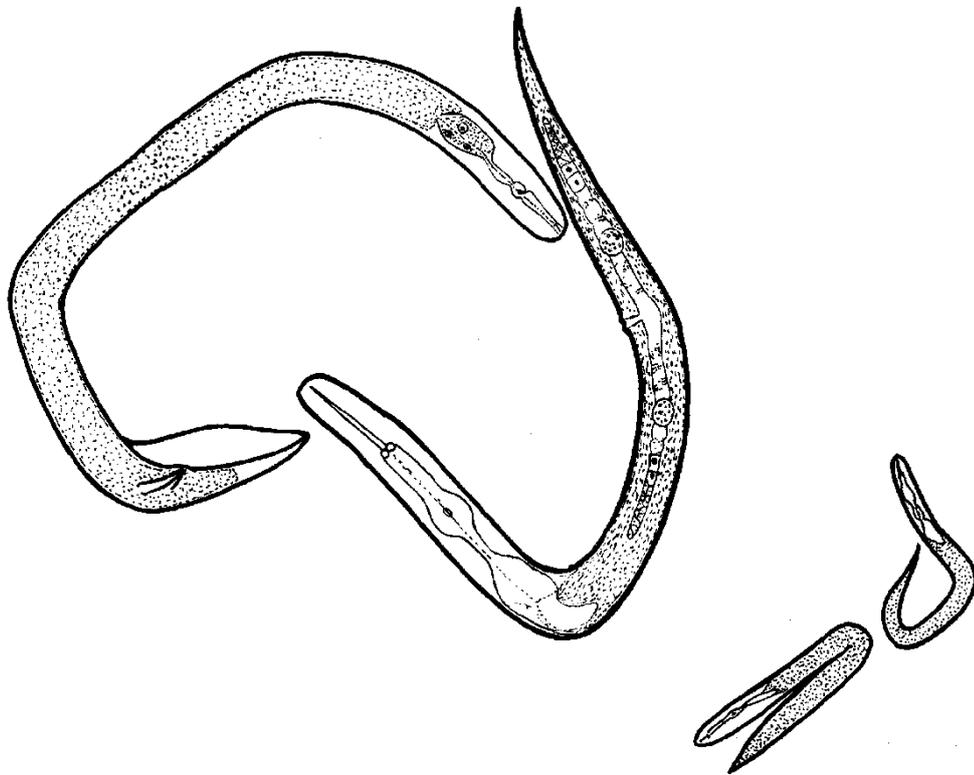


# AUSTRALASIAN NEMATODOLOGY NEWSLETTER



**Published by:**

**Australasian  
Association of  
Nematologists**

**VOLUME 22 NO. 1**

**JANUARY 2011**

# From the Editor

Thank you to all those who made contributions to this newsletter.

## July Issue

The deadline for the July issue will be mid June 2011. I will notify you a month in advance so please have your material ready once again.

*Kerrie Davies*

## Contacts

Dr Mike Hodda  
President, Australasian Association of Nematologists  
CSIRO Ecosystem Science  
GPO Box 1700  
CANBERRA ACT 2601  
Tel: (02) 6246 4371  
Fax: (02) 6246 4000  
Email: [mike.hodda@csiro.au](mailto:mike.hodda@csiro.au)

Dr Sarah Collins  
Secretary, Australasian Association of Nematologists  
Department of Agriculture and Food  
Locked bag 4  
Bentley Delivery Centre WA 6983  
Tel: (08) 9368 3333  
Fax: (08) 9474 2840  
Email: [sarah.collins@agric.wa.gov.au](mailto:sarah.collins@agric.wa.gov.au)

Dr Vivien Vanstone  
Treasurer, Australasian Association of Nematologists  
Department of Agriculture and Food  
Locked bag 4  
Bentley Delivery Centre WA 6983  
Tel: (08) 9368 3141  
Fax: (08) 9474 2840  
Email: [vivien.vanstone@agric.wa.gov.au](mailto:vivien.vanstone@agric.wa.gov.au)

Dr Kerrie Davies  
Editor, Australasian Nematology Newsletter  
School of Agriculture Food and Wine  
Waite Campus  
University of Adelaide SA 5005  
Tel: (08) 8303 7255  
Fax: (08) 8379 4095  
Email: [kerrie.davies@adelaide.edu.au](mailto:kerrie.davies@adelaide.edu.au)

# Association News

## FROM THE PRESIDENT

AAN is affiliated with the Australasian Plant Pathology Society as a special interest group, and as such we have always held a workshop and short meeting in association with their biennial meeting. The next APPS meeting will be held in Darwin on 26-29 April 2011, and the nematology workshop will be on the first day. The theme for the workshop is "Nematology in the Asia-Pacific: new and old threats to plants". We will be looking at the local situation regarding pest nematodes around Darwin in the morning, then talking about tropical nematology in the local region in the afternoon. We hope to have speakers from Fiji, Indonesia, China and other countries in the region to compare experiences with those from tropical Australia. There will, of course, be a general meeting of AAN and a dinner afterwards. Contact Mike or Barry Conde for more details. Thanks to Barry for acting as local organiser. See elsewhere for the formal invitation.

This meeting is held in conjunction with the Asian Conference on Plant Pathology. Traditionally, AAN has met at APPS, but with ASDS meetings becoming more frequent, fewer members attend any one meeting. After some discussion, we decided to hold one of the "traditional" nematology workshops in association with APPS, but I welcome thoughts on how and when it is best to hold nematology meetings for the majority of members (see agenda for the general meeting). With my other travels and commitments, I could not attend the ASDS meeting last year, even though I would have liked to. The attendance at the last APPS was also lower than normal, so it may be appropriate for the nematologists to decide to meet at one venue or the other.

While on the subject of meetings, remember that we now have the Australasian Nematology Support Fund to support travel to international and national meetings. The first round of grants received one application, which was going to be funded until the applicant received all the required funds from another source. So the funds have been added to the pool, and larger grants will be available next time. Students can apply to this fund at any time for assistance in attending Australian or international conferences. An application need only state what you want, what you will do at the meeting, and if there are any other funds available.

On the international front, you may be aware that AAN is part of the International Federation of Nematology Societies (IFNS). The main tasks of this organisation are to select the site for the International Congress of Nematology, and to organise a web site which links all nematology societies and resources. In Brisbane, Frida Decraemer was elected President, taking over from the wonderful job done by David Chitwood. Frida is an extremely active President, and in addition to choosing the site of the next international congress, has been investigating the formal legal position of the IFNS. At the moment, IFNS has the same legal status as AAN: that is, none. It has been an entirely voluntary organisation operating on an honour system, with no funds. (In the latter aspect it is different from AAN because we do have some funds.) This informal status causes problems if a society wants to do anything involving money or contracts, so Frida has been exploring the options, and has arrived at the point where, if IFNS is to be formally constituted, the member societies need to agree. To this end she has written a more detailed report which is elsewhere in this newsletter.

For what it is worth, I think that AAN has been down this road, and we seem to be doing adequately without formal incorporation (except for SICN which was incorporated as a

separate body), so I intend to vote for AAN appropriately. (We have 1 vote of 22 for IFNS.) However, if anyone has strong opinions on this, then please let me know soon, so that we can vote differently should we decide it is in the interests of AAN and the broader nematology community to do so.

*Mike Hodda*

## **Australasian Association of Nematologists - GENERAL MEETING**

6 pm, 27 April 2011, Darwin (Venue TBA)

### DRAFT AGENDA

Apologies

Minutes of Previous Meeting

Issues Arising

President's Report

Secretary's Report

Treasurer's Report

Newsletter Editor's Report

Australasian Nematology Support Fund

Election of officers

Next Nematology Workshop

Future meetings of AAN

Any other business

If there are any other items for the agenda, please contact the Secretary, Sarah Collins, at the address listed at the front. Despite the length of the above list, we hope each item will take only a few minutes.

The general meeting will be followed by a dinner at which less formal discussion can continue.

# Regional News

## NEWS FROM CANBERRA

Abdul Gafur is continuing his PhD candidature at ANU and for the past 2 years has been hunting for every specimen of *Radopholus* to be found. After locating fixed specimens of most species in the genus, he has been trying to re-collect as many as possible to obtain molecular samples, but has so far been unable to re-collect many species, despite several visits to the type localities. If anyone has found any *Radopholus* other than *R. similis* recently, Abdul would love to hear from you.

In the meantime, he has decided to expand the scope of his project to include all the Family Pratylenchidae, using specimens collected for Mike and other's past projects.

Sunil has completed his PhD confirmation seminar, held at CSIRO Ecosystem Sciences in October and recently had a glimpse of the Australian countryside on his trip to Wagga Wagga for the Post Graduate Symposium at Charles Sturt University. Currently he is analysing the nematode distribution, host range and identification data to determine exotic species which are of potential biosecurity significance. He is also looking at state-wide distribution of nematode species in order to determine potential targets for internal quarantine. A classic example is *Ditylenchus dipsaci* which has approximately 30 different pathotypes with differing pathogenicities and currently only three races are recorded from Australia hence pathotype specific quarantine measures may be needed for selected species. Meanwhile for other nematodes, screening at genus or species level could be sufficient. Preliminary results from the project are interesting and there still many pieces yet to be solved in the nematode biosecurity puzzle.

Natalie Banks, a technician at ANIC, has been doing a part-time project on the dispersal of Potato Cyst Nematodes, and has completed a final draft of a paper comparing rates of dispersal using different assumptions in different countries. The paper should be submitted by the time this report goes to press. Natalie is to be commended for her work in this area. Although she has yet to be converted to nematology, her recognition that they can be good subjects for studies of biosecurity (her real area of interest), shows that there may be hope yet. She is aiming to publish the paper in the journal *Phytopathology*.

Mike has been working furiously on a project on using genomic tools for the assessing soil communities, including all soil organisms, not just nematodes. Despite the promise of the technology, there are still many hurdles in going from genes to populations, including sampling, identification and interpretation. One of the major hurdles is actually finding a species corresponding to a sequence. Although we have many sequences from many economic pests, most free-living species and lesser pests remain unsequenced, making their molecular identification even more problematic than the identification of the many morphologically undescribed species.

Mike has also been continuing his checklisting project for the Australian Faunal Directory, checking the nomenclature of all nematodes of Australia (free-living and parasitic on invertebrates or vertebrates; marine, freshwater and terrestrial). Mike has started with the plant parasites, and will be proceeding through the marine nematodes, freshwater species and

invertebrate parasites before tackling the vertebrate parasites last. Mike had a long discussion with Ian Beveridge, from The University of Melbourne, and David Spratt, formerly of CSIRO, in regards to this latter group when they were in Canberra in December.

Joining the team part-time for the checklisting project is Debbie Jennings, formerly with a number of museums and scientific institutions in South Africa. Debbie has never worked with nematodes before, but has a great deal of experience in taxonomy of bigger, less interesting things.

In September, Mike attended a workshop on Potato Cyst Nematodes at Harper Adams University College (UK). The level of interest in PCN in the country was shown by a contingent of 3 from Australia (Mike, Sarah Collins & Dolf De Boer). We saw some very impressive work on rapid and cheap diagnostics, on suppression using non-host rotations, and Western Australia's declaration of area freedom from PCN (YAY Vivien and Sarah).

After a brief visit to the Mike's old stamping ground at The Natural History Museum in London, Mike attended the European Society of Nematologists meeting in Vienna. There he co-chaired the Ecology, Biodiversity and Evolution sessions with Hans Helder, where the various attempts at molecular ecology were presented and compared with more traditional approaches. His conclusion at the end of the sessions: "we are moving closer to being able to integrate the different strengths of the traditional and molecular methods to tackle ecological, management and identification problems that were previously intractable, and this is very exciting". There were also 2 sessions on biosecurity Mike presented a paper co-authored with Sunil and Natalie on nematode biosecurity which was well received. A highlight of this paper was the observation that of over 600 species of plant parasitic nematodes causing economic damage somewhere in the world, less than 20% of these were present in many major countries. For Australia, the figure was less than 10%, so the interest in biosecurity and quarantine of nematodes is definitely justified.

Other interesting sessions at the conference were on emerging pests (species of *Meloidogyne* and *Pratylenchus*), and interactions between nematodes and other disease organisms of plants.

*Mike Hodda & Sunil Singh*

## NEWS FROM QUEENSLAND

### **Brisbane, DEEDI**

The tropical and sub-tropical Nematology team in Queensland has had another busy and rewarding year. New projects have commenced and some older favorites have been successfully completed.

The team is working on various research projects involving plant-parasitic nematodes – one being the "Managing the nematode threat" project on root-knot nematodes ably described by Vivien Vanstone in the last edition of this newsletter. We have been heavily involved for many years in soil health research and have current projects on soil health in bananas and vegetables. We have been developing biological indicators of soil health, and as free-living nematodes respond quickly to changes in the food supply or environmental condition, they can be particularly useful in this area of research. Both Tony Pattison and Jenny Cobon have continued to develop their skills in the identification of free-living nematodes and are using them as indicators of the biological condition of the soil.

In January, Jenny Cobon travelled to Scotland on an OCPPO scholarship to study the morphological and molecular identification of *Globodera rostochiensis* and *G. pallida* (Potato Cyst Nematodes). She was instructed by Dr Jon Pickup and his team at SASA and Dr Vivian Blok and her team at SCRI. Jenny has submitted her diagnostic protocol to Office of the Chief Plant Protection Officer and given a presentation on her studies at a DEEDI/APPS seminar day in Brisbane. It was an invaluable experience to learn the diagnostic techniques required to identify this pest of significant quarantine importance from world experts and to have the hands-on experience with this pest that could only be gained outside Australia.

Like all of us, Wayne O'Neill also has non-Nematology projects to occupy his time. He works on other soil health related projects including an ACIAR project –“Integrated crop production of bananas in Indonesia and Australia”, which focuses on soil health in relation to *Fusarium* wilt of banana. This project enabled Wayne and Tony Pattison to travel to Indonesia this year and Wayne to attend the ISHS-ProMusa symposium in China where he gave a presentation on Panama disease in Australia and the diversity and distribution of the pathogen in Indonesia.

Tony recently attended the ACORBAT meeting in Colombia in November where he gave a presentation entitled “Using healthy soil to suppress soil borne diseases and sustain banana production”. Tony recently submitted his PhD thesis. A summary of his thesis “The importance of the antagonistic potential in the management of populations of plant-parasitic nematodes in banana (*Musa* AAA) as influenced by agronomic factors” appears later in this newsletter.

Tony's growing team in South Johnstone now includes Leanne Forsyth and Tegan Kukulies (non-nematode lovers) working on *Fusarium* and soil health projects. Leanne and Tony recently travelled to Indonesia for further capacity building/skill exchange with soil health project partners and Tegan has just received first class honors from James Cook University for her honors project on “Integration of organic matter into banana plantations in north Queensland: the effects on soil properties”.

Jenny recently travelled to Kiribati and Fiji on a soil health scoping study and identified *Pratylenchus coffeae* of taro in Fiji.

Our team in Brisbane now includes Timothy Shuey who will work on all nematode-related projects as well as our soil health projects. Tim has settled in well and is making a good contribution to the team.

Wayne and Jenny were kept very busy in 2010 with the discovery of *Aphelenchoides fragariae* in strawberry crops in Queensland's main strawberry growing region on the Sunshine Coast. Plant samples received by us in our diagnostic lab had symptoms including stunting with tight aggregation of crowns (some with a necrotic apical tip) and stunted petioles and flower stalks. Flowers on short stalks commonly had aborted or partly aborted floral parts. *Aphelenchoides fragariae* was found to be the cause of the problem, rather than *A. besseyi*, which has in the past caused similar disease episodes in Queensland strawberries. We have since undertaken numerous surveys, pathogenicity studies, nematicide trials and work is continuing on the etiology of the disease and any potential control measures for runner and fruit growers. This work will be presented at APPS in Darwin in 2011.

6ASDS was a resounding success in 2010. Graham Stirling ably headed the organizing committee which included Jenny and Wayne. The event was held at Twin Waters in August and was attended by over 120 delegates. Good luck to the Western Australians in 2012. We hear that you'll be using the same event coordinator – Sally Brown – an excellent choice!!!!

The Brisbane team has moved from our beloved Indooroopilly site where we had been ensconced for some 40 years. We have relocated to state of the art facilities known as the EcoSciences Precinct, Boggo Rd. These facilities have been built on the site of the previous, and infamous, state maximum security prison, so lots of jokes are flying around about returning the state's worst people back to where they belong. The heritage listed façade of the old prison remains intact, with the new building that houses several work units of DEEDI, DERM and CSIRO built just behind. There will be over 1000 staff when the facility is fully occupied, and many are not too sure they'll want to work late into the night with the possibility of ghosts (and probably not friendly ones!!!!) lurking around. All phone numbers and fax lines have changed, but email access remains the same (firstname.lastname@deedi.qld.gov.au)

*Jennifer Cobon*

### **Leslie Research Centre, DEEDI, Queensland**

Our nematode group (Kirsty Owen, Tim Clewett, Jason Sheedy and John Thompson) ran a well attended field day in early October 2010. Highlights of the day included a 2-year winter crop rotation experiment and the National Variety Test trials and a segment on the local TV news. In the crop rotation trial, the susceptibility of the first year crops (including faba bean, chickpea, wheat and even some barley cultivars) clearly reduced the yield potential of the intolerant wheat planted in the second year of the experiment. In contrast, the performance of tolerant wheat cultivars in the same crop rotation trial (and in the NVT trials) provided a good demonstration of the usefulness of choosing tolerant wheat cultivars. The drought-breaking Queensland rain has caused some headaches at harvest at one site and abundant yields at another trial site.

We have continued our campaign to raise awareness about root-lesion nematodes in the northern grain region and spoke at four GRDC Research Updates in the region in August 2010. We have been invited to speak at six more Updates in February-March 2011 and we will continue to raise awareness about root-lesion nematodes in towns we haven't visited before and provide more details about management strategies. The northern panel of GRDC are keen to make more growers and advisors in the region aware of root-lesion nematodes and have engaged Econnect Communications (see news item from Robbie Mitchell).

Nikki Seymour and Graham Stirling (Biological Crop Protection) have started a new GRDC-funded project - "Biological suppression of root-lesion nematodes in grain-growing soils". Yujuan (Jady) Li is also working on the project. In December, Graham gave a seminar at the Leslie Research Centre on "Biological suppression of *Pratylenchus*: what have we learned from studies in sugarcane; preliminary observations in grain-growing soils and where to from here?"

John Thompson and Nikki Seymour have a new paper *in press* – Inheritance of resistance to root-lesion nematode (*Pratylenchus thornei*) in wheat landraces and cultivars from the West Asia and North Africa (WANA) region. *Crop and Pasture Science*.

Ros Reen, Tim Clewett and John Thompson are looking forward to attending the APPS/ACPP conference in April and catching up with AAN members at the nematology workshop.

*Kirsty Owen*

## NEWS FROM SOUTH AUSTRALIA

### The University of Adelaide

In November, Lisnawita (Ita) arrived in Adelaide to work with Kerrie Davies. She is a nematologist from North Sumatra University, Medan, Indonesia, and is supported by a scholarship from the Indonesian Government's Program of Academic Recharge. Ita will be in Australia for 3 months, until mid-February. She is studying *Fergusobia* nematodes and the galls they induce, and is describing a new species from *Eucalyptus tereticornis*. Back home, she will be examining local *Syzygium*, a known host plant in India and Australia, for fergusobiid galls.

Katherine Linsell is now finalising experiments and beginning to write up her work for her PhD on 'Genetic and physiological characterisation of resistance to root lesion nematode *Pratylenchus* sp. in wheat'. The overall objectives of this project are to identify the genetic loci and closely linked markers for the *P. thornei* gene(s) in the partially resistant Sokoll/Krichauff population and to describe the resistance response.

*Kerrie Davies*

## NEWS FROM WESTERN AUSTRALIA

### Murdoch University

In 2010, there were two major stands of work:

#### 1. Murdoch University

Staff: Mike Jones, Reetinder Gill, Susan Phillip; PhD students: Matt Tan, Joanne Tan, Paul Nichol; M.Phil Student: Meenu Singh; Visiting Student: Revi (Temasek Polytechnic, Singapore).

University R&D focussed on the following areas:

- Australia India Strategic Research Fund (with Uma Rao, IARI, New Delhi) – the work focusses on Using Next Generation Sequencing of the transcriptomes of root lesion nematodes – the transcriptomes of two RLN species have been sequenced using both Roche 454 FLX and Illumina platforms. With Paul Nichol, Reet is undertaking the bioinformatics to assemble transcripts and annotate them. Similar work is ongoing in India, with cereal cyst nematode (*H. avenae*) as the target organism. This will provide a basis to genes required for parasitism with those of root-knot and other cyst nematodes.
- Susan Phillip continued the work of Zhaohui Wang, who left to take up a project leader position in Syngenta's new Biotechnology Lab in Beijing. In this work we demonstrated that it is possible to reduce infection by RKNs by targeting giant cells for destruction.
- Joanne Tan is working with root lesion nematodes to study the process of gene silencing. She has found that, under appropriate conditions, RLNs will take up dsRNA ('soaking' experiments) and inhibit target genes. She has found that two such genes, which affect nematode movement, can disrupt movement, and that a gene construct prepared from one species can also work on a different species, which suggests that broad spectrum synthetic resistance genes can be developed using this approach.

- Matt Tan, supported by the CRC for National Plant Biosecurity, has compared DNA and protein based diagnostics (eg MALDI-TOF mass spectrometry) for nematode identification, working on root lesion, cyst and root-knot nematodes. He is now using 2D gel electrophoresis to compare protein patterns from different nematodes.
- Meenu completed her M. Phil. and then developed a system using *Caenorhabditis elegans* to screen a series of new compounds for possible nematicidal activity.

The group collaborates closely with Vivien Vanstone and colleagues at DAFWA – Vivien provides the practical viewpoint and advice on handling root-lesion nematodes. She also co-supervises PhD student Matt Tan.

## 2. Nemgenix Pty Ltd

Staff: CEO – Dr Sean Hird, Mike Jones, John Fosu-Nyarko, Vaughan Agrez, Doug Chamberlain, Perry Francisco, Motiul Quader, Jamie Ong.

Nemgenix is a Murdoch University spin-out company based at the SABC Murdoch University. It was successful in raising funds from Venture Capital and a series of granting agencies to work on developing novel nematode resistance traits. It has a number of target crops. For example, having established a wheat transgenic pipeline, transgenic lines with enhanced resistance to root lesion nematodes have been generated. It has established a partnership with a major company to apply its work.

**Meetings, awards and travel:** Joanne Tan won the ‘Best student presentation’ at the APPS meeting held at DAFWA in her presentation of her work, and Matt Tan won the Best Poster Presentation (Plants) at the Combined Biological Sciences Meeting, Perth.

Mike Jones was the only Australian representative at the EU COST Action 872 meeting in Lisbon, where he presented current work. He also was invited by the Academy of Sciences to represent Australia at a COST Domain Meeting and submit a report – this provided a fascinating insight into the working of the EU and its internal politics! Mike also attended the International Cereal Cyst Nematode Initiative Workshop and the ESN meeting in Vienna. He also gave the keynote talk at a meeting at Kakatiya University in India (Biotechnology: A Global Scenario) and celebrated Devali on a 3 acre farm in Andhra Pradesh – avoid the buffalo milk curd is his advice.

Although not nematode oriented, Mike was also invited to be part of the review panel for the Centre of Excellence in Biotechnology, King Saud University, Riyadh, Saudi Arabia.

*Mike Jones*

# International News

First of all, Dr Rosa Manzanilla, the Vice-president, Safia Siddiqi, the Secretary and myself, your IFNS President wish you and the nematology society you represent a successful 2011. As last year, I want to provide you an update on the activities of IFNS realized in 2010.

The main activity concerned the organization of the selection process for the next congress site. By 1 March 2010, five societies (CSPN, JNS, NSI, NSSA, SON) had submitted a well documented proposal. On 4 March a first voting round was launched. It resulted in an equal number of highest votes for JPN and NSSA. **A second round in April had a winner:** the Nematology Society of Southern Africa will organize the 6<sup>th</sup> International Nematology Congress. I hereby wish to thank all councillors of the different societies for their commitment to vote and this within the proposed time schedule. It was a very exciting election and for me a real pleasure to organize it.

A second activity dealt with IFNS Constitution. At the end of 2009, all councillors received a ballot with the question if: 1) we should keep IFNS without legal status, with or without rectifying the draft of the constitution as a kind of working agreement, or 2) should IFNS go for a legal status as an international non-profit organization with tax-exempt status and keep the Federation registered in the US or go for another country. The response to the ballot was very low; no other suggestions were made except for the very kind proposal by Dr Rodrigo Rodriguez-Kabana to help us as an US citizen to explore what are the steps to be taken and the costs to go for a legal non-profit tax exempt status. The Attorney T. A. Bush of Sirote & Permutt of Birmingham, Alabama contacted by Rod informed us that the easiest way to proceed is to incorporate IFNS and be a C3 (tax exempt) organization. This requires formalization regarding rules and regulations pertaining among others, as to who is going to control the monies and financial activities and estimated budgets, if any (so far IFNS did not have finances). Most of this is already written or implied in the proposed constitution and would not be difficult to do. If we decide to give IFNS tax exempt status, then a mechanism will have to be developed to maintain objectivity, continuity and independence of the funds from the rest of the IFNS.

The estimated cost of the legal work will be U.S. \$4000 - 5000 plus \$700 user's fee. The cost will depend on how much information and work we can provide the Attorney and how much he has to actually do.

- Why go for a legal status? For example, last September, Safia Siddiqi had a big problem to renew the domain name [www.ifns.org](http://www.ifns.org) because IFNS is a not-for-profit scientific society that has not been officially registered nor has a business license.
- So far, we did experience problems but all is founded on good will and mutual confidence, respect and commitment since there are no binding statutes/rules.
- IFNS an officially registered society could make it easier to obtain financial support from official institutions and avoid possible communication problems or others by applying the Constitution.

Therefore, I would ask for your cooperation to give your opinion by filling in the new ballot.

A third activity concerns the IFNS website.

- So far the website has been regularly updated for announcements of nematology symposia and congresses.
- An update on new books published in 2010 will soon be uploaded.
- An update on Obituaries is ready to be uploaded.
- We will start with “*Well-known nematologists, role models for the science*” with a page and illustration on de Man (The Netherlands) based on his Biography by G. Karssen.
- The lay-out of a poster of IFNS has been developed but I need an original of the symbol of your society or pictures

As a fourth activity we have started our contacts with the Nematology Society of Southern Africa in order to discuss all matters with respect to the 6<sup>th</sup> Congress. A first meeting was held with Driekie, Mieke, Alex and Shaun in September 2010 in Rothamsted Research (UK). Rosa and I are going to attend the next NSSA symposium in May 2011 to continue activities related to the organization and scientific programme of the next Congress.

NEEDED from YOU:

- Provide us pictures of any nematode related subject so that every month we can update the home page of the website
- Send us information on nematology meetings, new job possibilities, interesting results, worrying developments,..
- Provide us an original of symbol of your society for the poster
- Send us the ballot

FORTHCOMING

According to the working rules of IFNS, art 4 on Structure of Organisation of the draft of the constitution: “At the end of 3 years, the same officers are re-elected or replaced by new officers in case of non-confidence voting or resignation for an additional 3 years. A quorum of Councillors from at least 5 Societies can request a vote of confidence for any officers anytime during their term.” I have contacted David Chitwood to organize this election. He has accepted this and will contact all councillors.

*Roza Manzanilla*  
*Vice-President*

*Wilfrida Decraemer*  
*President of IFNS*

# Research

## PLANT PARASITIC NEMATODES AND CONTROL WITH CHITIN FORMULATION IN PALM OIL PLANTATION AT SOUTH SUMATRA, INDONESIA

*Mulawarman*

Department of Plant Pests and Diseases, Faculty of Agriculture, Sriwijaya University, Jl. Palembang-Prabumulih Km.32, Inderalaya, Palembang 30662

Tel. 062-711-580663, Fax. 062-711-580663, Email: Mulaunsri@yahoo.co.id

Plant parasitic nematodes have limited agriculture productivity world-wide, especially in South Sumatra, Indonesia. Recently, the use of nematicide to control plant nematodes has caused human health and environment problems. The objective of the research was to investigate the effectivity of a chitin formulation in controlling plant parasitic nematodes in South Sumatra Palm Oil Plantation. Chitin has been investigated elsewhere in control programs.

The work began with collection and identification of the plant parasitic nematodes in the rhizosphere of the South Sumatra Palm Oil Plantation and ranking of the abundant nematodes. The plant parasitic nematodes were dominated by *Rotylenchus buxophilus* and *Xiphinema diversicaudatum*. The latter nematodes belong to a group which can vector plant viruses and hence it is hoped that they can be controlled with a chitin formulation.

The raw materials from which chitin can be obtained (shrimp and crab shells) are readily available in South Sumatra. They were obtained as waste from fish markets and seafood restaurants, and the chitin was prepared for this research by the deproteinase process. Given the volumes of waste available, use of chitin in soil amelioration would be economically viable.

Rhizosphere soil with abundant and known numbers of *R. buxophilus* and *X. diversicaudatum* was treated with a chitin formulation consisting of chitin, neem, and urea 250 g, or Furandan 150g. Each test plot was about 2 sq meters. Controls were without chitin formulation or nematicide. The *R. buxophilus* population was suppressed by chitin formulation 250g - 46.8 nematodes; Furandan 150g - 55.8 nematodes; and without chitin formulation or nematicide - 116 nematodes. Numbers of *X. diversicaudatum* were also suppressed: chitin formulation 250g - 44.6 nematodes; Furandan 150g - 53.2 nematodes; and without chitin formulation or nematicide; 112.2 nematodes. The chitin formulation could suppress the plant parasitic nematode population. The treatment may increase the number of antagonist microbia of plant parasitic nematodes.

## ECOFRIENDLY MEASURES FOR THE MANAGEMENT ROOT KNOT DISEASE IN RICE CAUSED BY *MELOIDOGYNE GRAMINICOLA*

*Anamika, Sobita Simon and Firdoos Ahmad*

Department of Plant Pathology, A.A.I., D.U., Allahabad, U.P., India

anamika\_bhu01@rediffmail.com

Rice is an important staple food crop of developing countries like India which is associated with nematodes causing incipient etiology and losses in yield. The world wide annual yield loss due to plant parasitic nematodes in rice ranges from 10-25%. Among plant parasitic nematodes, *Meloidogyne graminicola* is wide-spread in the rice growing area of Allahabad (U.P., India) and is considered the most economically important next to *Hirschmanniella* species (Rice root nematode). *Meloidogyne graminicola* causes root knot disease, one of the most destructive diseases in rice crops. This disease can be managed through biological control. The main purpose of the control is to suppress the inoculation load of the target pathogen to a level which would not cause potential economic loss in the crop. Pesticides, fungicides or nematicides are useful tools in agriculture but may cause gradual degradation of an ecosystem. Considering the harmful effects of these chemicals, an alternative approach is to use biological control of plant diseases. Limited work is being reported under integrated pest management. There has been considerable work in the field of biocontrol of *Meloidogyne* spp. causing root knot disease, leading to the introduction of various biological agents.

*Trichoderma harzianum* causes reduction in inoculum density of pathogens or parasites in its active or dormant state. One or more organisms may be naturally present or may be encouraged through manipulation of the environment.

The use of bacteria for management of plant disease and yield improvement has increased steadily. *Pseudomonas fluorescens* has emerged as the largest and potentially most promising group of plant growth promoting rhizobacteria (PGPR) involved in the biological control of plant disease. PGPR play a role in biological control by increasing populations of antagonists, increasing the vigour of the plants and inducing systemic resistance.

In the experiments reported here, it was concluded that among different control methods applied as seed and seedling treatments on *M. graminicola* of rice, T1 [*T. harzianum* @ 4 gm ( $4 \times 10^7$  cfu)/ kg] as seed treatment and T2 [*P. fluorescens* @ 4gm ( $4 \times 10^7$  cfu)/ kg] as seedling treatment were highly effective in reducing the numbers of *M. graminicola* and enhancing plant growth parameters. Significantly higher numbers were present in untreated plots.

Nematophagous fungi may be saprophytic in soil or decaying plant materials and may prey on saprophytic as well as plant parasitic nematodes. When populations of nematodes increase in soil, their natural enemies like nematophagous fungi are activated to parasitize and kill them and consequently cause a decline in nematode population. This may reflect a natural balance and buffering capacity of soil that does not allow development of a single dominant species; rather it maintains a good balance of soil biodiversity. The capturing efficiency of the predacious fungi may be influenced by the environmental condition and nature of the soil.

The presence of organic matter and nematodes is necessary to maintain the bio-diversity of the soil and increase the propagules of predacious fungi.

Amending the soil with application of mass cultures of two nematophagous fungi (*Arthrobotrys oligospora* and *Dactylaria eudermata*) reduced the number of root galls caused by *M. graminicola* by 86.9% and 81.1%, of females by 94.2% and 91.7%, of eggs + juveniles by 93.6% and 92.1%, respectively. The mass culture of these fungi increased the plant growth and shoot length by 41.9% and 38.8%, root length by 44.6% and 41.8%, fresh weight of shoot by 61.1% and 58.7%, and fresh weight of root by 24.3% and 22.5%, respectively over that of plants grown in nematode infested soil.

The use of commercial neem products e.g., Neemraj and Achook led to significant reduction in nematode populations in soil and increased the growth parameters as compared to the control.

Use of chemical pesticides viz. chloropyriphos 25 E.C and carboforan 3G at higher doses, i.e. 3 litre a.i./ha chloropyriphos and 4 kg. a.i./ha carboforan, led to a maximum decline in numbers of *M. graminicola* in soil as compared to the control.

Figure: Rice root knot caused by *Meloidogyne graminicola*



## STUDY ON BIOLOGY AND LIFE CYCLE OF RICE ROOT NEMATODE

*Anamika, Sobita Simon and Geeta Bhandari*

Department of Plant Pathology, A.A.I., D.U., Allahabad, U.P., India

Rice root nematode *Hirschmanniella* sp. is considered the most important nematode pest for rice on a global basis including India. The common name rice root nematode is derived from its specificity to rice and root parasitism. *Hirschmanniella* spp. are cosmopolitan in distribution; but *H. oryzae* is most widely distributed. In India, *Hirschmanniella oryzae* are encountered throughout the country in all rice growing areas. Rice root nematode infests 58% of the world's rice fields, causing 25% yield losses (Hollis and Kesboonruing, 1984).

*Hirschmanniella* spp. are migratory endoparasites. All the juvenile stages and adults (both male and females) are infective and penetrate roots just behind the root cap. Entry points made by one nematode facilitate the penetration of others through the same point. After penetration, nematodes move freely in the cortical cells. Soon after females start laying eggs which are scattered within the cortex. After hatching the juveniles continue migrating and feeding within the root and develop further. One life cycle is usually completed in a month. Generally 2-4 generations are completed in one crop season. The juveniles and adults can emerge into the soil in search of new roots. Nematodes attain peak population at the time of flowering.

The above ground symptoms are reddish brown discolouring of leaves and stunted growth of plants in patches. The infested plant roots show discolouration. Sometimes the symptoms are confused with nutrient deficiency. In general, there is arrested growth, poor tillering, reduced number of earheads and grain weight. On the roots, initial necrosis intensifies and by the time crop matures, the entire root system appears brownish and reduced in size.

Under North Indian conditions where rice/wheat rotation is followed, populations of *Hirschmanniella oryzae* reduce during the winter (rabi) season due to non-availability of hosts and low temperatures uncondusive for nematode activity. A small proportion of juveniles and adults survive through the winter season in rice stubbles. Alternate hosts were examined in experimental rice fields and the most preferred was *Echinochloa crusgalli* followed by *Eleusine indica* and *Cyprus iria*. Management of this disease is mainly through summer ploughing, destruction of stubble and crop rotation with non-host crops.



(a)

(b)

Figure: Penetration of rice root nematode in behind root cap of rice roots

# Reports

## CEREAL CYST NEMATODE BIOLOGY AND MANAGEMENT WORKSHOP

24<sup>th</sup> September 2010, Austria

(workshop following 30<sup>th</sup> International European Society of Nematologists Meeting)

*Julie Nicol*

CIMMYT, Ankara, Turkey

**Background:** This meeting was a follow up to the successful 1<sup>st</sup> International Cereal Cyst Nematode Workshop held in Turkey in 2009 where there were 60 participants from 20 countries. The organising committee for this meeting included colleagues from CIMMYT Turkey, Belgium (ILVO), UK (SCRI), China (CAAS) and Iran (IRIPP) (**Appendix 1**). The key purpose of this meeting was to:

- a) give European colleagues interested in CCN an opportunity to join the meeting
- b) update new information since the meeting in 2009
- c) discuss technical aspects of related work
- d) identify opportunities for collaborative initiatives

**Participants** – 28 participants from 14 countries attended the workshop (**Appendix 2**). A formal preparation of abstracts was prepared (**Appendix 3**) which was published in the proceedings from the International ESN meeting. Two contributions were not included here as colleagues were unable to produce the abstract in time – including a status of CCN in Russia and Central Asia (Dr Mikhail Pridannikov) and a molecular presentation from Australia (Prof Mike Jones).

**After the presentations an open discussion was chaired by Dr Florian Grundler and Prof Richard Sikora and the following summary notes and action steps were recorded:**

### **Methods should be standardized**

CCN resistance screening method

Evaluation of Resistance

Field Sampling

*Action group:* Zahra Maafi (lead), Julie Nicol, Ricardo Hildago, John Lewis (subject to confirmation)

Suggestion that 20% of the samples should also be checked for RLN.

### **Use of molecular tools**

Nematode aspects – diagnostics and phylogeny, genomic sequencing for discovery of R gene(s)?

Plant aspects – transgenics, RNAi, identification of genes of resistance (MAS)

Host parasite interactions and mechanisms – Bonn and BOKU have interest

Map the CCN genome - India (Uma Roa/Australia)

Mapping the wheat genome – almost completed

Should make sequence data base – Bonn to follow

Molecular ring test should be organised and coordinated

*Action group* : Leiven W (Belgium –subject to confirmation) and Bonn

*Participants* : Bonn, ILVO (Belgium), China (CASS Beijing + other), Iran (Maafi), India (Umaro –subject to confirmation), Australia (Jones – subject to confirmation), Turkey (Toktay), Tunisia (Julie to check), Russia (Mikhail).



**Participants from the Cereal Cyst Nematode Biology and Management Workshop**

**24<sup>th</sup> September 2010, Austria**

## **Root Lesion Nematode – where does it fit (could we also potentially add Dryland Roots Rots – Fusarium and Bipolaris)**

Some of key groups working on CCN should get data on RLN. Suggestion that 20% of the samples should also be checked for RLN

*Action group* – Turkey/CIMMYT(Halil Toktay – Julie Nicol), China (Deliang Peng), India (AK Singh – subject to confirmation), Morocco (INRA - Abbad Anndalousi (INRA – Morocco – subject to confirmation), Tunisia (INRAT – Najoua Kachouri subject to confirmation), Algeria (Aissa Mokabli), Russia (Mikhail), Saudi Arabia (???), Jordan (??).

## **Drought Stress and Physiology Research**

*Action group:* - CIMMYT (Julie Nicol), Bonn (Grundler, Sikora, Christain?), Mike Jones (Australia)

Can we make hot spots for yield loss and the affects of climate change in the future

## **Soil Health – Conservation Agriculture**

Julie Nicol to follow up with National program partners the opportunities to use.

Should include rotation and tillage retention practices

Johannes Hallman may help with biodiversity – metagenomics – subject to confirmation

Countries using Wheat and Maize rotation need to determine the importance of Maize (China/India – other).

## **ICCNI (International Cereal Cyst Nematode Initiative) Committee Membership**

Iran and Bonn Uni Germany will join the existing group of ILVO (Belgium), Rothamsted (UK), CIMMYT (ICWIP), Turkey and China. There was no representation from INRA so the potential role of France is not clear in this ICCNI committee.

## **Resource Mobilization Committee - suggestions**

Collective summary of activities and holistic approach of work

Maximum 3 page update of what we know

Anyone who identifies funding options should send to Julie and she will circulate to the wider audience.

## **Training**

If you have specific training needs please inform Julie Nicol

## **Next meeting – 2<sup>nd</sup> International Cereal Cyst Nematode Workshop, China, October 2011**

Prof Deliang Peng offered to host a 3-day workshop next year in China. It is hoped more than 40 international colleagues will attend. Both Deliang and Julie will identify opportunities for donor support to enable our NARs partners to join. Deliang will circulate more information about this meeting in the near future.

## Appendix 1

The sessions for the one day workshop included:

1. Current global status of the distribution of CCN
2. The economic importance and population dynamics of CCN on wheat
3. Control strategies of CCN on wheat using host resistance
4. Control strategies of CCN other than host resistance
5. Use of molecular tools for research with CCN (such as pathogen diagnostics, phylogeny studies and host resistance).

The workshop was open to anyone interested in working on Cereal Cyst Nematode from both Europe and other regions of the world. Both applied and theoretical research were discussed with a view to enhanced collaboration between research partners.

### Cereal Cyst Nematode Biology and Management Workshop – List of Participants

Abdel Naser Elashry	Germany	elashry@uni-bonn.de
Alexander Schouten	Germany	aschout@uni-bonn.de
Alfonso Martinuz	Bonn?	martinuz@catie.ac.cr
Amer Dababat	CIMMYT Turkey	a.dababat@cgiar.org
Christer Magnusson	Norway	christer.magnusson@bioforsk.no
Christian Hillnhütter	Germany	christian.hillnhuetter@gmx.de
Daria Shumilina	Russia	dasha2409@yandex.ru
Deliang Peng	China	dlpeng@ippcaas.cn
Didem Saglam	CIMMYT Turkey	d.saglam@cgiar.org
Emre Evlice	Turkey	emreevlice@gmail.com
Fard Karimipour	Iran	karimipourfard@gmail.com
Fateh Toumi	Syria	fateh_t@hotmail.com
Florian M.W.Grundler	Germany	grundler@uni-bonn.de
Halil Toktay	Turkey	toktay@yahoo.com
Holger Bohlmann	Austria	holger.bohlmann@boku.ac.at
Hongmai Li	China	lihm@njau.edu.cn
İ. Halil Elekçioğlu	Turkey	halile@cu.edu.tr
Julie Nicol	CIMMYT	j.nicol@cgiar.org
Lieven Waegenberge	Belgium	lieven.waeyenberge@ilvo.vlaanderen.be
Mike Jones	Australia	m.jones@murdoch.edu.au
Mikhail Pridannikov	Russia	mikhail.pridannikov@yahoo.com
Mohamed Selim	Egypt	m_elwy76@yahoo.com
Mustafa İmren	Turkey	mustafa_imren@hotmail.com
Ricardo Holgado	Norway	ricardo.holgado@biotorsu.no
Richard Sikora	Bonn, CGIAR	rsikora@uni-bonn.de
Shahid Siddique	Germany	siddique@uni-bonn.de
Yuji Oka	Israel	okayuji@volcani.agri.gov.il
Zahra Tanha Maafi	Iran	tanhamaafi@yahoo.com

## Appendix 2: Abstracts from CCN Workshop –Vienna September 2010

### 1. An update: Current global status of Cereal Cyst Nematode (*Heterodera spp.*) research on wheat – opportunities and future needs

Nicol, J.M.<sup>1</sup> and Rivoal, R.<sup>2</sup>

<sup>1</sup>CIMMYT, ICARDACIMMYT Wheat Improvement Program, PO Box 39 Emek 06511 Ankara, Turkey.

<sup>2</sup>Biologie des Organismes et des Populations appliquée à la Protection des Plantes (BiO3P), UMR INRA/ENSAR, BP 35327, 35653 Le Rheu, France (retired).

The 1<sup>st</sup> Workshop of the International Cereal Cyst Nematodes Initiative was conducted in Turkey October 2009 which was represented with 60 participants from more than twenty countries providing clear evidence of the global importance of this Nematode on wheat (The proceedings can be downloaded at <http://www.spipm.cgiar.org/reports>). Historically research was initiated intensively developed from the 1970's on cereal cyst nematode (CCN) focused mainly on the species *Heterodera avenae* in Australia, North America, western Europe and Asia. Much later, other species were reported including *Heterodera latipons* in the Mediterranean, *Heterodera hordecalis* in northern Europe and *Heterodera filipjevi* predominately in eastern Europe. The present situation indicates the importance of Cereal Cyst Nematode across large areas including many countries within the region of North Africa, West Asia, Middle East, China, India, Australia and the Pacific North West of the USA. The three major species are reported with *H. avenae* and *H. latipons* having more essentially a Mediterranean distribution, whilst a less understood and described species *H. filipjevi* being frequently reported from continental climates and is expanding in its distribution. New reports particularly from China, Iran, Tunisia and USA have documented the economic importance of CCN. Research on the distribution and economic impact demonstrate that several of these species cause important losses especially under rain-fed conditions and limited irrigation as in Australia, western Asia, north Africa, China and Pacific Northwest of the USA. Biological cycles and temperature requirements for activity of juveniles differ according to the species and populations inside species and demonstrate different hatching schemes and adaptation process to climatic conditions and new work has now been reported on the less understood *H. filipjevi*. One of the major challenges to the work is the complex genetic relationships between populations and cereals in the host reactions with the characterisation of pathotypes of which the virulence must be known for the use of resistant cultivars. Although progress has been made in the molecular identification of species, rapid markers to differentiate species are still not available, and as research expands our knowledge of the diversity and distribution of CCN species and pathotypes, such as those in India and China. Combined morphologic and molecular data have improved knowledge about phylogeny of the CCN complex. Research on resistant and tolerant cultivars have identified effective sources of resistance against some species and pathotypes, with many of these molecularly characterised with known confirmed *Cre* genes which has enabled the use of MAS (Marker Assisted Selection) strategies in some breeding programs. Wheat lines (germplasm) developed by CIMMYT, ICARDA and Turkey in collaboration with National Program partners offers a chance to combine sources of resistance. Distribution of this wheat germplasm to partner countries to better understand the usability of genetic resistance. Exploring parasitism genes offers opportunities to understand resistance with view to using genetic transformation. Futuristic work on both mapping the CCN Genome would offer excellent opportunities to further explore resistance and phylogeny. Further integrated control options have been established with various rotation schemes, seed treatment and the use of resistant cultivars constitute the most appropriate management options to maintain the population densities below the damaging levels. The International Cereal Cyst Nematode Initiative which began in 2007 and has offered the opportunity for a new set of investigators, bringing with it the opportunity to bring research partners together with research complementarities. This one day Cereal Cyst Nematode Biology and Management Workshop

offers the opportunity for researchers to present some of these new findings. Acknowledgements : contributions from colleagues in China (Prof D Peng, Prof H. Li, Dr S Chen), India (Dr AK Singh, Dr DJ Kaur), Iran (Dr ZT Maafi), Tunisia (Dr Namouchi-Kachouri), Turkey (Dr N Bolat, Prof H Elekcioglu, Dr H Toktay, Dr Elif Sahin, Mr M Imren, Ms D Saglam), Belgium (Dr L Waeyenberge, Dr N Viaene, Mr F Toumi), CIMMYT (Dr AA Dababat, Dr Y Manes), ICARDA (Dr F. Ogbonnaya), USA (Prof R. Simley) and Australia (Dr J Lewis, Dr IT Riley). The published proceedings Riley IT, Nicol JM, Dababat AA eds (2009) 'Cereal cyst nematodes: status, research and outlook.' (CIMMYT: Ankara, Turkey).

## 2. Current progress of the cereal cyst nematodes (*Heterodera avenae* and *H.filipjevi*) on wheat in China

Peng, D.<sup>1</sup>, Nicol, J.M.<sup>2</sup>, Huang, W.<sup>1</sup>, Chen, S.<sup>3</sup>, Zheng, J.<sup>4</sup> and Li, H.<sup>5</sup>

<sup>1</sup>State Key Laboratory for Biology of Plant Diseases and Insect Pests, Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing 100193, China.

<sup>2</sup>CIMMYT, ICARDA-CIMMYT Wheat Improvement Program, Turkey Office, Ankara, Turkey.

<sup>3</sup>Institute of Plant Protection, Hebei Academy of Agriculture and Forestry Sciences, Sciences, Baoding 071000, China.

<sup>4</sup>Department of Plant Pathology, Nanjing Agricultural University, Nanjing 210095, China.

<sup>5</sup>Institute of Biotechnology, Zhejiang University, Hangzhou 310029; 2Horticulture and Forestry Bureau of Jinzhong, Shanxi, Jinzhong 030600, China.

China is the world's largest producer of wheat >120Mt pa and average 4t/ha yield. A summary of the work on cereal cyst nematode (CCN) in China was reported in the Proceedings of the 1<sup>st</sup> Workshop of the International Cereal Cyst Nematodes Initiative conducted in Turkey October 2009. Up to 2009, *Heterodera avenae* was confirmed in 12 wheat growing provinces in central, north, northwest and east China, covering a large area of the Chinese bread basket, including both the optimal (high rainfall/irrigation) and rainfed (dryland) wheat production systems. A new record of CCN is reported from Ningxia province with frequency of 59.8% from 80 samples. Yield losses have been confirmed from several important wheat growing provinces including Henan, Hebei and Anhui and based on survey data ~22% of the wheat growing area of China may potentially suffer losses of 10% yield, i.e., an estimated loss of 1.2Mt of wheat. A report based on both morphological and molecular identification has confirmed that four populations of CCN from Henan Province are *H. filipjevi*. Molecular data of rDNA-ITS and D2D3 region were submitted to GenBank, being a new record of *H. filipjevi* in China (Peng DL., *et al.*, Plant Disease, 2010 in press). The pathotypes of thirteen CCN populations from various provinces using the international standard differential hosts and a common local cultivar Wenmai 19, indicate that most of the 12 populations differ from the described pathotypes previously in the worldwide. Only one population belonging to Xingyang was found to be similar to the Xushui pathotype (Ha43) (Yuan, *et al.*, 2010; Li & Chen, 2010, unpub. data). The resistance of 116 wheat varieties and lines from CIMMYT and Turkey to CCN Baoding population was screened and evaluated in a growth chamber. The results showed that there are 15 highly resistant, 15 moderately resistant, 36 moderately and 50 highly susceptible varieties (Chen, 2010). Whilst most Chinese cultivars are susceptible a few appear to provide some level of resistance. The amplification the rDNA-ITS and D2/D3 regions of cereal cyst nematode populations collected from Gansu, Henan and Anhui provinces in China along with UPGMA revealed alignment with related *H. avenae* populations (AY148382) and related species *H. aucklandica* (DQ328688) and *H. australis* (AY148395). Further work on 22 scored fragments from Gansu population were obtained with 9 restriction enzymes. Digested PCR amplified ITS of *H. avenae* populations collected from Gansu province suggest 7 of these populations might be type C of *H. avenae* and different from type A of European populations and type B of Indian populations. Three new parasitism genes including *Ha-cbp-1*(GenBank accession GQ178086), *Ha-pel-1*(GenBank accession GQ998895) and *Ha-pel-2* (GenBank accession GU138156) were

cloned by RACE kit based on homologous cloning method and appear to be associated with *H. avenae* resistance. Work is continuing on all aspects of this research.

### 3. Longterm studies on the Cereal Cyst Nematode with winter wheat (*Heterodera filipjevi*) and new research with *H. avenae* in Turkey: International collaboration with regional implications

Elekcioglu I.H., Nicol, J.M.<sup>2,6</sup>, Bolat, N.<sup>3</sup>, Şahin, E.<sup>1,2</sup>, Toktay, H.<sup>4</sup>, Yorgancilar, A.<sup>3</sup>, Braun, H.J.<sup>2</sup>, Imren, M.<sup>1,4</sup>, Ocal, A.<sup>1,4</sup>, Yorgancilar, O.<sup>3</sup>, Yildirim, A.F.<sup>4</sup>, Kilinc, A.T.<sup>3</sup>, Dababat, A.A.<sup>2</sup>, Erginbas, G.<sup>2</sup>

<sup>1</sup>Çukurova University, Faculty of Agriculture, Department of Plant Protection, Balcalı Adana, Turkey.

<sup>2</sup>CIMMYT, ICARDA-CIMMYT Wheat Improvement Program, Ankara, Turkey.

<sup>3</sup>Anatolian Agricultural Research Institute, Eskisehir, Turkey.

<sup>4</sup>Plant Protection Research Institute Adana and Diyarbakir, Turkey.

Preliminary work started in 1993 established the widespread distribution of cereal nematodes in both East Mediterranean region and Central Anatolia of Turkey. In 2001, a new joint initiative was established between CIMMYT International, the Turkish Ministry of Agriculture and Çukurova University to work on the 5Mha of rained winter wheat on the Central Anatolian plateau (CAP). CCN was found in 78% of some 300 soil samples collected from the CAP. All were identified morphologically and molecularly to be *H. filipjevi* and only one sample was *H. latipons*. Hatching behaviour of *H. filipjevi* was examined under in-vitro conditions which revealed the highest hatching percentage was obtained with 15, 10 and 5C treatments at 94, 92 and 75% respectively. Hatching was also significantly increases with temperature change from, 5C to 20C and from 10C to 20C at a rate of 49 and 42% (Sahin *et al.*, 2009). Yield loss trials were conducted over two years (2002/03 and 03/04) in two locations on the Central Anatolian Plateau (Haymana and Cifeler) under natural field conditions. The average yield loss (comparing treated and untreated nematicide plots) in 2002/03 was 20% for Cifteler and 36% in Haymana across the twelve varieties. The two commonly cultivated winter cereals Bezostaya and Gerek suffered high losses of 47 and 37%, respectively. Considering the drier year initial (2002/03) densities between 5-10 *H. filipjevi* eggs/g can be considered economically damaged (causing >10% yield loss). A long-term rotation trial established on the Haymana (Ankara) Ikizce experimental station since 1975 has been monitored in March 2003-05 for populations of cyst nematode under the different rotational combinations including chickpea, fallow, safflower, spring lentil, sunflower, wheat, vetch/barley mix, winter lentil and winter vetch. The fallow/wheat rotation for cyst nematode was found to show no difference when compared to wheat/wheat rotation, inferring fallowing does not significantly reduce cyst populations. In order of hosting ability for cyst nematode over the three years vetch/barley>wheat>fallow>chickpea, sunflower, spring lentil, winter lentil, winter vetch>safflower. From this study fallowing is considered not effective method to reduce cyst populations. Barley should be avoided under cyst populations are high (Elekcioglu *et al.* 2004). The most effective method to screen for resistance is under controlled greenhouse conditions with mass culture of CCN. More than 1,000 national and international winter and spring wheat lines were screened in order to determine their resistance against *H. filipjevi* local isolate TK1 from Haymana. Repeated screening has identified 28 lines to have resistance as good as or better than the known control lines (Nicol *et al.* 2009). Of the known published *Cre* genes, *CreR*, *Cre1* and the Milan VPM source - *Cre5* (also possibly *Cre2* and *Cre6* genes) was found to provide some level of moderate resistance, while others tested (*Cre3* and *Cre8*) were found to be ineffective. Many of the lines identified are in high yielding adapted spring and winter wheats, and several represent Turkish released cultivars. Further work is needed to validate this resistance under natural field conditions. The effectiveness of designated *Cre* genes depends on both the species of CCN and pathotype. Several specific CCN wheat nurseries have been distributed to more than 10 collaborator countries to confirm their usability. New work has

begun in the SE Anatolian region (provinces of Gaziantep, Kilis, Şanlıurfa, Mardin) which is both a facultative and spring wheat environment. Preliminary surveys indicate 53% of the 128 soil samples have CCN, however species include *H. avenae* and *H. latipons*. Similar work will be conducted in these regions for CCN with an emphasis on host resistance to these species. Preliminary evidence screening CIMMYT International spring wheat germplasm suggests many promising sources of resistance against local isolate *H. avenae* from Gaziantep.

#### 4. Current progress on Cereal Cyst Nematodes (*Heterodera filipjevi*, *H. avenae* type B and *H. latipons*) of importance on wheat in Iran

Maafi, Z.T<sup>1</sup>, Ahmadi, A.<sup>2</sup>, Hajihassani, A.<sup>3</sup>, Karimipour, H.<sup>4</sup> and Nicol, J.M.<sup>5</sup>

<sup>1</sup> Iranian Research Institute of Plant Protection, P.O. Box 1454, 19395 Tehran, Iran

<sup>2</sup> Plant Protection Department, Agricultural Research and Natural Resources Centre of Khuzestan, P.O. Box 613353341, Ahvaz, Iran

<sup>3</sup> Agricultural Research and Natural Resources Centre of Markazi

<sup>4</sup> Plant Protection Department, Agricultural and Natural Resources Research Center of Isfahan, P.O. Box 81785-199, Isfahan, Iran

<sup>5</sup> International Maize and Wheat Improvement Centre, P.O. Box 39, Emek 06511 Ankara, Turkey

Five species of cyst-forming nematodes belonging to the *Heterodera avenae* group were identified from cereal fields and grasslands in Iran, *H. avenae*, *H. filipjevi*, *H. hordecalis*, *H. latipons*, and *Heterodera* sp., of which *H. filipjevi* and *H. latipons* are the prevalent species in cereal fields respectively (Tanha Maafi *et al.* 2007). *H. latipons* is mainly widespread in west and central parts; whilst *H. filipjevi* distributed in most wheat growing regions throughout the country, *H. avenae* type B as the third prevalent species is found in west and south-west provinces i.e. Kermanshah, Ilam, Khuzestan where contains most of the rain-fed wheat culture system (Tanha Maafi *et al.*, 2009). A national project is underway in Khuzestan, Lorestan, Isfahan, Khorasan and Golestan provinces to study the occurrence, distribution and population density of CCN are being investigated in these regions using both morphological and molecular tools. In Khuzestan province both *H. filipjevi* and *H. avenae* were found on 37 and 35% of wheat and barley fields with population densities (mean(range)) from 18 (1-103) cysts/100g and 437 (0-1400) eggs and J2's/100g soil for wheat. In Isfahan province only *H. filipjevi* was identified being found in 53% of wheat and barley fields with population densities on wheat from 15 cysts/100g and 1658 (8-29700) eggs and J2's/100g soil. In many fields CCN was found in combination with other root diseases. In Lorestan province *H. filipjevi* and *H. latipons* were found in 52% of soil samples and mean population was 14(1-48) cysts/100g soil. In Golestan province *H. filipjevi* was identified to in 11% of soil samples with average cyst population of 58 cysts/100g soil. The biology and life cycle of two species of CCN *H. filipjevi* and *H. latipons* were studied on the winter wheat and *H. avenae* type B on spring wheat which revealed one generation per growing season with *H. filipjevi* and *H. latipons* completing its life cycle around 150 days compared with 90 days for *H. avenae* type B on spring wheat (Hajihassani *et al.*, 2010a; Ahmadi and Tanha Maafi, 2010). Yield loss trials were conducted in 2006 and 2007 to determine the impact of different initial population levels (0- 20 eggs and second-stage juveniles (J2) (g soil)<sup>-1</sup> of *H. filipjevi* and *H. latipons* on wheat revealed significant grain yield losses of 48 and 55% and regression models (Hajihassani *et al.*, 2010b, Hajihassani *et al.*, 2010c). Further yield loss studies of wheat under natural field densities (10 eggs and juveniles *H. filipjevi*/g soil) results in reductions in 53% reductions in grain yield using nematicide as a control. Further yield loss studies are underway in a number of provinces. The multiplication and assessment of resistance to CCN under *in vitro* conditions will soon begin with both National and International wheat lines from the joint CIMMYT-Turkey collaboration.

## 5. New distribution report of cereal cyst nematode on wheat in Shandong province of China

*Zhao, H.<sup>1</sup>, Yang, Y-y<sup>1</sup>, Peng D.<sup>2</sup>, Lio, F.<sup>3</sup>*

<sup>1</sup>College of Agronomy and Plant Protection, Qingdao Agricultural University, Qingdao 266109, China

<sup>2</sup>State Key Laboratory for Biology of Plant Diseases and Insect Pests, Institute of Plant Protection, CAAS, Beijing, 100193, China

<sup>3</sup>Plant Protection College, Shandong Agricultural University, Taian 271018, China

In May and June of 2009, 115 samples of wheat roots and rhizosphere soils were collected from 61 counties/districts of 13 regional cities in Shandong province, China. Cereal cyst nematodes (CCN) were found in 66.1% samples (76 samples) with the cyst densities ranged from 0~326 cysts per 200 ml soil. CCN occurred in 47 counties/cities/districts belong to the 11 regional cities of Zaozhuang, Liaocheng, Qingdao, Jining, Heze, Jinan, Dezhou, Bingzhou, Dongying, Zibo and Weifang. The CCN population densities differed greatly between the regions. Occurrence ratio were much high in the first five cities, with average cyst densities being high in the Liaocheng, Qingdao, Dezhou, Heze and Dongying. CCN cysts were not detected in 14 counties/districts of the 13 regional cities and were not found in all the 15 samples from 7 counties/districts of Rizhao and Linyi regional cities. The results suggest spreading of CCN over long distance may be associated with the trans-regional operations of wheat combine harvesters and the water flowing of large rivers. The CCN cyst densities were high enough to cause heavy damage to wheat production in some wheat blocks in Shandong province.

## 6. Investigation of Cereal Cyst Nematode in Ningxia Province of China

*Huang, W-K., Ye, W-X, Wang G-F, Long, H-B, Peng, D.*

State Key Laboratory for Biology of Plant Diseases and Insect Pests, Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing 100193, China

The cereal cyst nematode (CCN) is the most important pathogen of the wheat and other cereals in China and has adverse effect on the quality and production of wheat. The occurrence and distribution of this nematode was investigated by random sampling in 16 cities of five regions of Ningxia province. CCN was identified with the morphological and metrological examination, revealing that it was distributed in most areas of five regions of Ningxia province and the detection rate of 59.8 percent in all samples. The highest cyst number existed in Yinchuan city and the lowest cyst number existed in Shizuishan city. The amount of eggs per cyst in Guyuan, Zhongwei, Wuzhong and Qingtongxia city were significantly higher than that of Shizuishan city. The species of CCN was identified as *Heterodera avenae* Woll. based on morphological/metrological examination. These findings suggest that CCN may be an important biotic constraint in this region and requires further study.

## 7. The life cycle of *Heterodera avenae* type B on Chamran spring wheat cultivar under field conditions in Southwest of Iran

*Ahmadi, A.R.<sup>1</sup> and Tanha Maafi, Z.<sup>2</sup>*

<sup>1</sup>Plant Protection Department, Agricultural Research and Natural Resources Centre of Khuzestan, P.O. Box 613353341, Ahvaz, Iran

<sup>2</sup>Iranian Research Institute of Plant Protection, P.O. Box 1454-19395 Tehran, Iran

Two species of cereal cyst nematodes, *Heterodera avenae* and *H. filipjevi* are widely spread in wheat growing areas in Southwest of Iran. The life cycle of *H. avenae* type B was studied on

Chamran spring wheat cultivar under rain-fed field conditions in Behbahan agricultural research station during growing season 2009-2010. The first observation of the second stage juvenile (J2) was in late November with population density of 467 J2s/ 250 g soil and soil temperature of 20°C. The penetration of J2 was observed in early to mid December with soil temperatures 14.5°C and 15°C respectively. The highest population density of J2s with 1967 J2s/250 g soil was observed in early January with soil temperature of 12.3°C. The third and fourth stages were observed in mid January with soil temperatures 11.3°C and 12 °C respectively. White young females were visible on the root surface in the late January and soil temperature of 12.1°C, however, the highest population density of white females was observed from mid April till mid June with soil temperatures of 15.9°C and 15.8°C respectively. The male individuals were found in the first week of March till mid May with soil temperatures of 15.8°C and 20.6°C respectively. The brown cysts were appeared one month after white female occurrence with soil temperature 14.1°C. The results showed that *H. avenae* type B developed one generation per growing season in field conditions.

#### **8. Three years studies on distribution and population density of *Heterodera filipjevi* in cereal fields of Isfahan province, Iran**

*Karimipour Fard, H.<sup>1</sup> and Z. Tanha Maafi, Z.<sup>2</sup>*

<sup>1</sup>Agricultural and Natural Resources Research Center of Isfahan, P.O. Box 81785-199, Isfahan, Iran

<sup>2</sup>Iranian Research Institute of Plant Protection, P.O. Box 1454-19395, Tehran, Iran

Five species of cyst-forming nematodes belonging to the *Heterodera avenae* group were identified from cereal fields and grasslands in Iran. In this study the distribution and population density of cereal cyst nematodes, *Heterodera filipjevi* was surveyed in Isfahan province. Two hundred and six composite soil and root samples were randomly taken from different wheat and barley fields during 2008-2010. The root samples were examined under stereomicroscope to observe the mature female and cyst. The soil samples were processed by the Fenwick can method for cyst extraction. To determine the population densities of CCN in infested soil, the filled and semi filled cysts were counted, then crushed in a glass crusher and the numbers of second stage juveniles (J2) and eggs were determined. *H. filipjevi* was identified based on morphological and morphometric characters of J2 and vulval cone region of cysts, and rDNA-RFLP. *H. filipjevi* was found in 53.3% of soil samples, ranging from 51.7 and 68.7% in wheat and barley fields respectively. The mean number of cyst in wheat soil samples was determined 15 cysts/200 g soil, and the mean population of eggs and J2s was counted 1658 ranging from 8 in Meymeh region to 29700 in Zavareh region, whereas in barley it was 19 cysts/200 g soil and the population of eggs and J2s ranged from 63 in Aran Va Bidgol region to 16058 in Zavareh region with the mean of 2009. The white females and brown cysts were observed on root samples of *Phalaris minor* and *Avena ludoviciana* as well as wheat and barley. In some fields root rot diseases were found in combination with cereal cyst nematode.

## 9. Influence of a seed treatment on the infection and development of the cereal cyst nematode '*Heterodera filipjevi*' on susceptible and moderately resistant wheat germplasm

Dababat, A.A.<sup>1</sup>, Pariyar, S.R.<sup>2</sup>, Nicol, J.M.<sup>1</sup>, Erginbas, G.<sup>1</sup>, Wartin, C.<sup>3</sup>, Klix, M.<sup>3</sup>, Bolat, N.<sup>4</sup> and Sikora, R. A.<sup>2</sup>

<sup>1</sup>CIMMYT (International Maize and Wheat Improvement Centre), ICARDA CIMMYT Wheat Improvement Program, Ankara, Turkey

<sup>2</sup>Soil - Ecosystem Phytopathology & Nematology, University of Bonn, Germany.

<sup>3</sup>Syngenta Crop Protection AG, WRO 1004.6.65 Schwarzwaldallee 215, P.O. Box, CH-4002 Basel, Switzerland.

<sup>4</sup>Anatolian Agricultural Research Institute, Eskisehir, Turkey.

This study was carried out with a fungicide used as a seed coating for soil-borne pathogen control to determine its efficacy in reducing infection of the sedentary cereal cyst nematode *Heterodera filipjevi* on moderately resistant and susceptible winter wheat germplasm under the controlled conditions in ATEAM, Eskisehir, Turkey. The three different susceptible wheat germplasm (Seri, Bezostaya & Gerek) and three moderately resistant wheat germplasm (F130L 1.12/ATTILA, Katea & Sonmez) were tested at different fungicide concentrations of 25 g ai/100 kg seed, 50 g ai/100 kg and 100 g ai/100 kg seeds. The fungicide was applied as standard seed coating used to control damping off fungi. The moderately resistant germplasm gave a significant reduction in nematode reproduction in terms of cyst number per root system when compared to the susceptible germplasm. A significant reduction in the number of *H. filipjevi* was observed on the susceptible germplasm (Seri and Bezostaya) when the seed was treated with the fungicide. Neither shoot height, shoot weight nor root length were significantly increased by the seed treatment when compared with control. The greatest increase in shoot height was detected on the fungicide treated and moderately resistant germplasm. No phytotoxicity symptoms were observed on the plant at any of the fungicide concentrations used. The results seem to show that the fungicide reduced plant susceptibility to *H. filipjevi* infection by mechanisms still to be identified.

## 10. Pathotypes of cereal cyst nematodes in the Hebei province in China and evaluation of the resistance of wheat breeding lines to the nematodes

Li X., Ma J., Chen S.

Institute of Plant Protection, Hebei Academy of Agricultural and Forestry Sciences/IPM centre of Hebei Province, Baoding 071000, China

Cereal cyst nematode (CCN) is one of most important diseases affecting wheat production in Hebei. The occurrence of CCN was investigated at the time of wheat harvesting by extracting cysts from the soil. The results showed that CCN is widespread in Hebei province, except in the area in which spring wheat is grown, and that there is a trend for the number of cysts extracted from the soil to increase from the north to south and from east to west in Hebei. The pathotypes of the CCN populations from Baoding and Renqiu were tested with an international assortment of cereal cultivars and found to be different from all other pathotypes reported. The resistance of 116 wheat lines from CIMMYT to a local CCN population (Baoding) were evaluated in a growth chamber at 17-22°C. In the test each plant was inoculated with 1500 infective juveniles. The number of cysts from each cultivar was checked with a floating method after a two month incubation. Fifteen cultivars were highly resistant, 15 cultivars were moderately resistant, 36 cultivars were moderately susceptible and 50 cultivars were highly susceptible. Only three of the 26 cultivars which are known to be resistant to the Turkish population of *Heterodera filipjevi* were highly resistant to the local population. Effects of different temperature on the hatch of CCN collected from different period were evaluated in 24-well plates. The results showed that cold temperature treatment (5-15°C) is necessary for the CCN hatching. Most nematodes hatched in 6-7 weeks at 5-15°C. The CCN did not hatch at room temperature without

prior cold temperature treatment. CCN did not hatch at 28-30°C whether they had been pre-treated with a cold temperature or not. 20°C was found to be the best temperature for nematode hatch after cold treatment.

## 11. Preliminary survey of cereal cyst nematodes on wheat in Jiangsu province, China and analysis of the rDNA-ITS region from Jiangsu populations

Li H.<sup>1</sup>, Wang X.<sup>1</sup>, Pei S.<sup>1</sup>, Le X.<sup>1</sup>, Peng D.<sup>2</sup>

<sup>1</sup>Department of Plant Pathology, Nanjing Agricultural University, Nanjing 210095, China

<sup>2</sup>Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing 100193, China

The Cereal cyst nematode (CCN) was first reported from the Hubei province in the centre of China in 1989. Subsequently, it has been found in 11 provinces including Henan, Hebei, Anhui and Shandong provinces which are the top wheat producing areas in China. Jiangsu Province is located in East China and has a wheat growing area of approximately 2.3 Mha. A preliminary survey carried out during May of 2009 showed that Jiangsu was the 12<sup>th</sup> province with CCN. The wheat growing area of these 12 provinces represents about 80% of the total for China, around 20 Mha and CCN is therefore a serious problem for wheat production in China. In the survey, 76 samples with roots and soils were collected from 22 counties of 5 districts in Jiangsu province. The results of the nematode extraction and identification demonstrated that cereal cyst nematode was distributed widely in Xuzhou, Liangyungang, Suqian and Yancheng districts. The incidence rate was 35%. The CCN incidence rates in wheat growing areas of Peixian, Fengxian, Pizhou and Suining county from Xuzhou district were higher than 90%. The population densities detected from the fields of Huashan town (Fengxian), Hekou town (Peixian) and Suburb of Fengxian were higher than the other investigated fields, with average 109, 86 and 55 eggs/g soil respectively, which were much higher than those recognized as economically damaging to wheat. The ITS region of ribosomal DNA was amplified from 15 Jiangsu populations and the ITS-RFLP patterns revealed all populations with the same fragments after digestion with 8 restriction enzymes. The Jiangsu populations not only obtained the same patterns as the “B” type (Indian populations) by *AluI* and *RsaI* digestions, but also the “C” type by *HinfI* and *Tru9I* digestions which were specialized for Chinese populations. Phylogenetic trees constructed on basis of ITS sequences revealed that all of Jiangsu populations were grouped with the species from *Heterodera avenae* complex, which was one of the branches of the *H. avenae* group. Most of the Jiangsu populations were genetically closely related to the Russian and German populations of *H. pratensis*. The comparisons of ITS sequences from Jiangsu populations with other CCN populations from China and abroad revealed that rich genetic diversities were present in CCN populations from Jiangsu and other provinces in China.

## 12. Development of a species-specific duplex PCR to detect the cereal cyst nematode *Heterodera latipons*

Toumi F.<sup>1</sup>, Waeyenberge L.<sup>2</sup>, Dababat A.<sup>3</sup>, Nicol J.M.<sup>3</sup>, Viaene N.<sup>2</sup>, Kumarse N.<sup>4</sup>, Moens M.<sup>2,5</sup>

<sup>1</sup>Laboratory of Nematology, Ghent University, K.L. Ledeganckstraat 35, 9000 Ghent, Belgium

<sup>2</sup>Institute of Agricultural and Fisheries Research (ILVO), Burg. Van Gansberghelaan 96, 9820 Merelbeke, Belgium

<sup>3</sup>International Maize and Wheat Improvement Centre (CIMMYT), Ankara, Turkey

<sup>4</sup>International Centre for Agricultural Research in Dry Areas (ICARDA), Aleppo, Syria

<sup>5</sup>Laboratory for Agrozoology, Ghent University, Coupure links 653, 9000 Ghent, Belgium

Twenty four *Heterodera* populations were collected from wheat fields in Syria and Turkey and from existing cyst collections. DNA was extracted from one cyst of every population and the ITS region was amplified and sequenced. Comparison of these sequences with those available in Genbank (NCBI) showed/confirmed that the populations belonged to 9 species. Eleven populations were identified as *H. latipons*. Seven genes were selected to assess their

possibilities for designing species-specific primers for *H. latipons*. PCR products were obtained from actin, Hsp90 and tubulin genes. Weak or no PCR products were obtained from pectate lyase, annexin, chorismate mutase and aldolase. The obtained PCR products were purified, sequenced and aligned with *Heterodera* sequences of the corresponding genes available in Genbank (NCBI). The alignments were used to find DNA sites suitable for the design of species-specific primers for *H. latipons*. We were able to construct two species-specific primers within the actin gene. In addition, we constructed a reverse universal actin primer because the alignment revealed that the original primer primed on a polymorphic DNA-site. Several PCR assays with different combinations of the universal and species-specific actin primers were tested. One primer combination was retained and further combined with a universal 28S rDNA primer set which served as an internal control for the PCR. The ideal annealing temperature (gradient PCR), concentration of nucleotides and primers were determined. The optimized duplex PCR was subsequently tested on all samples. Results showed that a reliable and robust *H. latipons* species-specific duplex PCR was constructed. However, testing more populations and species originating from all over the world (specificity) as well as determining the lowest possible level of detection (sensitivity) is still needed.

### **13. Sequence and RFLP analysis of rDNA-ITS and 28S rDNA-D2D3 regions of *Heterodera avenae* on wheat from Gansu Province in China**

*Ye W.<sup>1,2</sup>, Xu B.<sup>1</sup>, Peng D.<sup>2</sup>, Huang W.<sup>2</sup>*

<sup>1</sup>Pratacultural College, Gansu Agricultural University, Lanzhou 730070, China

<sup>2</sup>State Key Laboratory for Biology of Plant Diseases and Insect Pests, Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing 100193, China

Cereal cyst nematode (CCN) is an important pathogen of cereal crops which is present in a number of wheat-growing areas in China. The 28S rDNA-D2/D3 and the rDNA-ITS regions of *Heterodera avenae* (CCN) populations collected from Gansu province, Anyang county of Henan province and Bangbu county of Anhui province in China were amplified. In each case a fragment of the 28S rDNA-D2/D3 region of approximately 780 bp and a fragment of the rDNA-ITS of approximately 1040 bp was amplified. The D2/D3 and ITS sequences amplified from the *H. avenae* populations from Gansu province, Anyang county of Henan province, Bangbu county of Anhui province in China were compared with the related species *H. aucklandica* (DQ328688), *H. australis* (AY148395) and *H. avenae* (AY148382) using UPGMA. These analyses showed that the nematodes from China were clustered in the same group with a close relationship. A total of 22 scored fragments were obtained with 9 restriction enzymes from digests of PCR amplified ITS of *Heterodera avenae* (CCN) populations collected from Gansu province in China. The 9 restriction enzymes produced restriction profiles that were identical for all the populations in Gansu province. Combination of the 28S rDNA-D2/D3 and the rDNA-ITS regions analysis and rDNA-ITS RFLP analysis showed that 7 populations of Gansu province might be type C of *H. avenae* and different from type A of European populations and type B of Indian populations.

### **14. Molecular cloning and sequence analysis of cellulose binding protein gene (*Ha-cbp-I*) from the cereal cyst nematode (*Heterodera avenae*)**

*Gu X.<sup>1,2</sup>, Peng D.<sup>1</sup>, Peng H.<sup>1,2</sup>, Long H.<sup>1</sup>, Wang G.<sup>1</sup>, Huang W.<sup>1</sup> and He Y.<sup>2</sup>*

<sup>1</sup>The State Key Laboratory for Biology of Disease and Insect Pests, Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing 100193, China

<sup>2</sup>Key Laboratory of Agricultural Biodiversity and Pests Control, Ministry of Education, Yunnan Agricultural University, Kunming 650201, China

The cereal cyst nematode, *Heterodera avenae*, is an important nematode pathogen of wheat in China. Cellulose binding proteins are important for parasitism and invasion by these plant

parasitic nematodes. The cDNA sequence of *Ha-cbp-1* (GenBank accession GQ178086 ) was cloned by RACE kit based on homologous cloning method. The results showed that the cDNA sequences of *Ha-cbp-1* contained an open reading frame, which encoding 131 amino acids with a predicted signal peptide sequence for secretion and a cellulose-binding domain. The gDNA sequence of *Ha-cbp-1* contained two introns with the length of 932 bp. The predicted HA-CBP-1 amino acid sequence had 60% identity and 75%-76% similarity with HS-CBP-1 and HG-CBP-1, cellulase binding proteins from *Heterodera schachtii*.

## **ADVISORS AND GROWERS HELP RAISE ROOT-LESION NEMATODE AWARENESS IN THE NORTHERN GRAIN REGION**

*Robbie Mitchell*

Econnect Communication Pty Ltd., PO Box 3734, South Brisbane Bc, Qld 4101

Root lesion nematodes are a 'sleeping issue' in the northern grain region according to the Grains Research and Development Corporation's (GRDC) northern panel chair, James Clark.

Costing unsuspecting northern grain growers \$47 million dollars a year in yield losses, GRDC contracted Econnect Communication to raise awareness about root-lesion nematodes with growers and advisors working in the region. Our plan has been to listen to what the researchers have to say, find out how much growers and advisors know and how best to communicate to them the importance of managing for nematodes.

Since July 2010, we have worked closely with Agri-Science Queensland (Department of Employment Economic Development and Innovation) researchers at the Leslie Research Centre in Toowoomba, Queensland, to draft a comprehensive communications strategy which targets farmers and advisors.

Before we started to get the message out there, we wanted to understand what level of awareness and knowledge farmers and advisors had. To do this, we drafted a survey and sent it to over 2,000 farmers and advisors in the region. Although it was a busy time for growers, we received over 100 responses, which have given us valuable information.

The responses will tell us how serious people think the situation is, which sub-regions require greater assistance, what information needs to be more clearly communicated and how best to communicate it.

To help consolidate the large amount of helpful information already out there, we will also create a root-lesion nematode website for growers and advisors to find out more about root-lesion nematodes, if paddocks in their area have root-lesion nematodes, how to spot crop symptoms and, most importantly, how to get their soil tested. We are currently building the website and hope to have it online by late February 2011.

We will publish the results of the survey on the website and in Issue 91 of next year's *Ground Cover* magazine.

For more information on the campaign, or if you would like a copy of the survey report, please don't hesitate to contact me on 07 3846 7111 or [robbie@econnect.com.au](mailto:robbie@econnect.com.au).

# Thesis Abstract

## **THE IMPORTANCE OF THE ANTAGONISTIC POTENTIAL IN THE MANAGEMENT OF POPULATIONS OF PLANT-PARASITIC NEMATODES IN BANANA (*MUSA* AAA) AS INFLUENCED BY AGRONOMIC FACTORS**

*Tony Pattison*

DEEDI – Agri-Science Queensland, PO Box 20, South Johnstone QLD 4859

Summary of thesis submitted in partial fulfilment of the requirements for the degree of Doctor of Philosophy, 2010.

*Radopholus similis* is a major obstacle to sustainable banana production around the world. Traditionally, the nematode has been managed through the use of nematicides, which have the potential to move into the local environment and pose a health risk to farm workers. However, in some soils plant-parasitic nematodes have been observed to have little impact on banana production. The difference in agricultural practices used on farms has been shown to have a significant impact on the soil environment. Therefore, a better understanding of how soil management practices impact on the soil environment may help to develop a greater understanding of soil properties that can lead to the suppression of plant-parasitic nematodes in banana plantations.

The use of organic amendments was investigated as one method that may stimulate organisms that are antagonistic to plant-parasitic nematodes. Nine different amendments; mill mud, mill ash (by-products from processing sugarcane), biosolids, municipal waste (MW) compost, banana residue, grass hay, legume hay, molasses and calcium silicate (CaSi) were applied in a glasshouse experiment. Significant suppression of *R. similis* occurred in soils amended with legume hay, grass hay, banana residue and mill mud relative to untreated soil. Furthermore, the applications of banana residue, grass hay, mill mud and MW compost were able to increase the nematode community structure index, indicating greater potential for suppression of plant-parasitic nematodes through predation.

Where additional organic matter was applied there was a decrease in the number of *R. similis* recovered from the roots of banana plants and an increase in the growth of banana plants. When cellulose and a mixture of carbon sources were applied there was also a decrease in the number of *R. similis*. The cellulose treatment increased the number of fungivores and omnivores in the soil, but resulted in a lower labile carbon level in the soil relative to the untreated soil. The suppression of plant-parasitic nematodes resulting from the addition of moderately degradable organic matter, like cellulose and grass hay, appeared to be the result of a combination of two factors; nematotoxic compounds produced in the early degradation of the organic matter, followed by an increase in nematode antagonists favoured by an increase in soil fungal activity.

A field experiment was established to determine the changes in soil properties following application of four soil amendments; compost, grass hay and by-products from sugar mills, mill mud and mill ash. The amendments were applied at the time of planting bananas and re-applied again 12 months later. At the termination of the experiment there was significant increase in bunch size in the mill ash treatment and decline in bunch size in the compost and mill mud treatments relative to the untreated plants. Furthermore, in the soil treated with organic matter there was an increase in labile C, the number of omnivorous nematodes and lower proportion of plant-parasitic nematodes relative to the untreated soil.

A study was implemented on 10 banana plantations in the north Queensland banana production area to determine differences in soil management, physical, chemical and nematode community properties using a multivariate statistical approach to determine factors that would demonstrate differences between plantations and suppression of plant-parasitic nematodes. A principal component analysis was able to explain 61% of the total variation between farms and identified the soil factors with the highest loadings as; the proportion of plant-parasitic nematodes, labile C, nitrate-N, and the number of fungal feeding nematodes in the soil community. A forward stepwise regression of the soil properties associated with the proportion of plant-parasitic nematodes in the soil community found that the ratio of labile C and nitrate-N in the soil and the diversity of nematodes was able to explain an adjusted 88.7% of the variation in the proportion of plant-parasitic nematodes in the soil. Farm practices that increase carbon inputs, manage nitrogen fertiliser applications and promote soil biodiversity, can increase soil labile C, reduce nitrate-N and increase nematode diversity leading to a suppression of plant-parasitic nematodes on banana plantations in Australia.

A survey of 21 banana plantations in Costa Rica measured soil chemical and physical properties as well as the nematode community composition. A principal component analysis using 34 soil variables was able to explain 71% of variation between plantations using the first five principle components. A banana bioassay of the soil from the 21 plantations, inoculated with *R. similis*, resulted in different populations of the nematode recovered from the root system. A forward stepwise regression of the number of nematodes recovered from the roots of bioassay plants with soil properties revealed that pH, structure index and Zn gave a significant multiple linear regression model, which was able to explain 79.2% of the variation in recovery of *R. similis* from the roots of bioassay plants. Furthermore, the correlation of soil pH with nematode diversity, the proportion of predatory and omnivorous nematodes and the structure of the nematode community suggested that pH was the factor limiting the biological suppression of *R. similis* in the Costa Rican banana plantations.

The development of soils capable of suppressing plant-parasitic nematodes requires and understanding of the inherent soil constraints in the farming system. In Australia, soil C appeared to be limiting, which was correlated to the activity of organisms antagonistic to plant-parasitic nematodes. Therefore, an over supply of nitrogen fertilisers altered the balance of carbon and nitrogen, resulting in a decrease in the activity of organisms antagonistic to plant-parasitic nematodes. The application of amendments that are high in C appeared to be able to induce suppression of plant-parasitic nematodes in bananas by developing a more favourable environment for antagonistic organisms. However, in Costa Rica low soil pH constrained the diversity of the soil nematode community, which corresponded to higher numbers of plant-parasitic nematodes in the roots of bananas. Management options to increase soil pH need to be investigated to determine if they contribute to the suppression of *R. similis* in Costa Rican banana plantations.

## Other News

### NEW PRODUCT FOR NEMATODE BIOCONTROL

from *IPMnet NEWS* #183, December 2010

A U.S. firm has announced a breakthrough for liquid culture production of *Pasteuria* spp., a group of naturally occurring soil bacteria known to specifically attack plant-parasitic nematodes. *Pasteuria* commercially grown in nematode hosts are currently used in limited amounts, but at prohibitively high costs for most agricultural applications, a situation, the firm says, is solved by their liquid culture innovation. *Pasteuria* forms endospores and therefore can be applied, packaged, and stored. The firm's first product has gained necessary governmental clearances, and registration applications are pending for additional products, according to their website [www.pasteuriabio.com](http://www.pasteuriabio.com).

Pasteuria Bioscience, Inc., 12085 Research Dr., Suite 185, Alachua, FL 32615, USA.  
[DDuncan@pasteuriabio.com](mailto:DDuncan@pasteuriabio.com). Voice: 1-386-462-0008, ext. 1.

Excerpted, with thanks, from the indicated website.



The 4<sup>th</sup> Asian Conference for Plant Pathology and the 18<sup>th</sup> Biennial Australasian Plant Pathology Society conference will be held in Darwin, NT from 26 – 29 April 2011. Early registration closes 21<sup>st</sup> January 2011.

Dr Peter Williamson  
Business Manager  
APPS Inc.  
[www.appsnet.org](http://www.appsnet.org)

The nematology workshop is as detailed below.

**Nematology in the Asia-Pacific: new and old threats to plants; conducted by Mike Hodda and Barry Conde on 26 April 2011.**

**Cost (pp) \$155. Minimum 20, maximum 35 people.**

In the morning we will tour vegetable farms in the Darwin area to view Root Knot Nematode problems in the Darwin area, an area far from other agricultural areas, yet with many of the same issues with nematodes. We will discuss what we can learn from this.

In the afternoon, there will be a symposium on nematology in the Asia-Pacific (China, Vietnam, Malaysia, Indonesia, Fiji, northern Australia and other places). There will be presentations from experts on the main tropical species (Root Knot Nematode, Burrowing Nematode and the Rice nematodes), as well as discussions of newly-identified threats, cooler climate nematodes in the tropics (eg Potato Cyst Nematode in highland areas of Indonesia), and nematode biosecurity and trade in the Asia-Pacific Region.

**Location:** Field trip then Charles Darwin University (Transport from/to conference venue provided).

**Duration:** One day, 08:30 – 17:00.

**Registration includes:** Morning tea, lunch and afternoon tea.

# NEMATODOLOGY IN A SCHOOL SCIENCE COURSE

John Sagun of the Middle School Science Department at Marrara Christian College in Darwin NT 0812 has been introducing his students to plant and soil nematodes. Attempts were made to extract them from soil and the rhizosphere of Gamba grass (*Andropogon gayanus*), a highly invasive weed in savannah land of the Northern Territory. They were hoping to find plant parasitic nematodes that could possibly be damaging to the root system of Gamba grass.

Photos were taken by Year 7 students. The poster below was created by Bronte Malin, a Year 9 student. It won 1<sup>st</sup> place for the scientific communication category in the NT's Young Scientist Award held at Charles Darwin University in 2010.

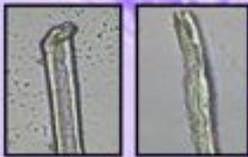
Congratulations to John for an imaginative course and for actually telling his students about nematodes! And congratulations to the students on their work. It is great to know that they are aware of some of the fascinating life that is under our feet.



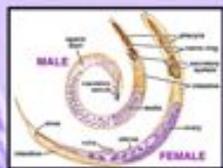
### Nematode Morphology and Biodiversity

Bronte Malin  
Marrara Christian College  
01/06/2010

**Nematode Morphology:**  
Possible Plant and Fungi Feeders



**Nematode Structure**



**Introduction:**  
Nematodes are said to be one of the most abundant multi-cellular organisms on earth and can be found in every habitat around the world (including polar regions). They come in many types, shapes and sizes and are very important as biological control methods in farming and agriculture.  
In this experiment we studied the biodiversity of nematodes found in the soil at Marrara Christian College and took photos of the nematode structure. The soil sample used came from a mildly moist garden bed in the courtyard.

**Variables:**  
Independent: — moisture in the soil — food available to nematodes  
Dependent: — number and biodiversity of nematodes

**Methods:**

- Using a spoon, place a small amount of soil into a cup.
- In the funnel, place the netting followed by the tissue and then the soil sample.
- Clamp the test tube and place the end of the funnel into the top of the test tube.
- Using the spray bottle, slowly spray the soil. The water will filter through the soil, tissue and netting and end up in the test tube.
- Take the test tube of water and place a drop of the water on a microscope slide, using the dropper. Place the slide cover over the top.
- Next, place the slide under the microscope and focus it.
- Look for any nematodes that are present.

**Results:**

A)

Type of Nematode	Present or Absent
Bacteria Feeder	Absent
Fungi Feeder and/or Plant Feeder	Present

B) Predicted Food Chain:

```

graph LR
    Fungi --> Fungi-Feeding-Nematode
    Bacteria --> Bacteria-Feeding-Nematode
    Plant-root --> Plant-Feeding-Nematode
    
```

**Discussion:**

The plant and fungi-feeders have a stylet in their mouth.

A) The techniques we used eventually proved to be successful, although it was hard for us to find the nematodes and it took a long time.  
 B) One of the mistakes we made was accidentally squirting water on our microscope slide. This affected what we could see. Another mistake we made was collecting soil that wasn't very moist and wasn't the best place to find nematodes.  
 C) We could have improved our method by collecting moist soil and also making sure our microscope was focused properly.  
 D) The only type of nematode that we found was the plant or fungi-feeder. This would most likely be because the soil came from around plants. The plant and/or fungi feeder has a stylet.  
 E) Studying nematodes benefits agricultural yields and farming by allowing people to understand the importance of nematodes as biological control agents.

**Conclusion:**  
In our investigation we only found minimal numbers of plant eating nematodes. The methods and techniques we used were reasonable. There was not really any biodiversity in our nematodes because we only found plant and/or fungi-feeders. Retrieving a moist soil sample is likely to have produced better results.






**Acknowledgement:** Thanks Mr. Sagun for helping us during this experiment.

**References:**  
<http://www.cmo.csiro.au/science/nematodes/introduction.html>  
<http://www.cals.ncsu.edu/course/en150/mooley/fall/nematode.jpg>

## SHORT COURSE

### NEMATODES IN CROPPING SYSTEMS - IDENTIFICATION AND TECHNIQUES

The next course is scheduled for Adelaide in late November/early December 2011. It will be run over 5 days.

If you are interested, please submit a non-binding expression of interest to Mike Hodda at CSIRO Entomology or Kerrie Davies at The University of Adelaide (addresses below).

As in previous presentations of this course, Kerrie Davies and Mike Hodda, the co-convenors, try to tailor the course to suit the needs of participants. We envisage the following.

The workshop will suit researchers and professionals working in agriculture, quarantine, green keeping, and soil biology, who need to understand the principles and practice of handling soil, plant and insect nematodes. It will provide hands-on experience in sampling, extraction, specimen preparation, culturing, diagnosis, and identification (including molecular techniques). There will be opportunity for interaction with experts in the field.

Participants should have a degree which includes biology, agriculture, or soil science or have appropriate work experience to undertake the workshop. Less experienced participants can be supplied with recommended reading material prior to the workshop.

Nematodes to be considered include:

*Meloidogyne Tylenchulus Heterodera Tylenchorhynchus*  
*Pratylenchus Morulaimus Ditylenchus Radopholus*  
*Anguina Bursaphelenchus Scutellonema Hemicycliophora*  
*Paratrichodorus Filenchus Xiphinema Tylodorus*  
*Aphelenchoides Heterorhabditis Helicotylenchus Steinernema*  
Rhabditida Mononchida Dorylaimida Areolaimida

Anticipated course cost is \$1760 (including GST). This includes all materials and a printed course manual.

Details of course content can be varied to suit the interests of the participants. Please contact the co-ordinators to discuss any specific needs or topics desired for inclusion.

Dr Mike Hodda  
CSIRO Ecosystem Science  
GPO Box 1700  
Canberra ACT 2601  
Phone: 02 6246 4371  
Fax: 02 6246 4000  
Email: [mike.hodda@csiro.au](mailto:mike.hodda@csiro.au)

or

Dr Kerrie Davies  
Plant Protection  
School of Agriculture Food and Wine  
The University of Adelaide,  
Waite Campus  
Glen Osmond SA 5064  
Phone: 08 8303 7255  
Fax: 08 8379 4095  
Email: [kerrie.davies@adelaide.edu.au](mailto:kerrie.davies@adelaide.edu.au)