

# **Report for 2003CA35B: Structure and seasonal changes of nematode communities from vernal pools (Santa Rosa Plateau)**

There are no reported publications resulting from this project.

Report Follows

## ***Structure and seasonal changes of nematode communities from vernal pools (Santa Rosa Plateau)***

### **Introduction**

Nematodes are diverse and abundant in soils and sediments, occupying a wide range of ecological roles that reflects the overall condition of the microbiological ecosystem. Our project constitutes the first study of nematode communities from vernal pools. It aims to provide the first ecological and taxonomic data from this fragile and biologically important habitat, through a combined morphological and molecular survey of two pools in the Santa Rosa Plateau Ecological Reserve (SRPER). These data will be analysed for the purposes of ecosystem health monitoring, and for possible occurrence of nematodes parasitizing the locally occurring endangered species of plants and fairy shrimp.

### **Research Program**

During the first year of this project, our activities consisted mainly of the training of undergraduate student helpers, and the collection of two sample series. State permits were approved by October 2002, but unfortunately the federal permit took until May 16<sup>th</sup> 2003 to arrive. The first sample series was collected on June 11<sup>th</sup> 2003, each sample consisted of five 200 ml cores taken randomly with a 6x2 inch auger in between plant stems (to make sure no plants were removed), within a 1 m<sup>2</sup> square. Limited physicochemical measurements (temperature, pH, conductivity, oxidation reduction potential) were taken on-site. Soil texture and moisture were manually and visually assessed on-site.

Two samples were collected from each of two vernal pools during each sampling series. The four resulting samples were taken to the nematology greenhouse for extraction of fairy shrimp cysts and nematodes, using sieving and decanting methods. In the following days and weeks, nematodes were processed for video microscopy and PCR as outlined in the original proposal. The remainder from each sample was fixed in hot formalin-glycerin 4:1, transferred to pure glycerin and 100 randomly picked individuals were mounted in permanent slides for identification.

The second sample series was collected on November 3<sup>rd</sup>, 2003 and processed as before, with two additions: water contents was determined by drying a subsample, and visual inspection of the root and plant debris fraction was followed by overnight extraction in a mist chamber.

### ***Genera and species identified to date***

The table below is a list of all nematode taxa isolated between March 2003 and February 2004 from SRPER. Genus identifications are largely complete and have yielded no fewer than 51 different genera. Species identifications are in progress, nine species were

identified to date and three of these are potentially new to science (confirmation requires literature from various obscure sources that are still being tracked down).

<i>Achromadora</i>	<i>Mesorhabditis</i>
<i>Acrobeloides</i>	<i>Microdorylaimus</i>
<i>Anaplectus</i> sp. cf. <i>granulosus</i>	<i>Monhystrella</i>
<i>Aphelenchoides</i>	<i>Neopsilenchus</i>
<i>Aphelenchus</i>	Nordiidae
<i>Aporcelaimellus</i>	Nygolaimidae
<i>Basiria</i>	<i>Nygolaimus</i>
<i>Boleodorus</i>	<i>Panagrolaimus</i>
Cephalobidae	<i>Paractinolaimus</i> new. sp.??
<i>Cephalobus</i> new sp.??	<i>Paraphanolaimus</i>
Chromadoridae	<i>Plectus</i>
<i>Coslenchus rhombus</i>	<i>Pratylenchus</i>
<i>Ditylenchus</i>	<i>Prismatolaimus</i>
<i>Dorylaimellus</i>	<i>Prodesmodora</i>
<i>Dorylaimus</i> new. sp.??	<i>Psilenchus</i>
<i>Enchodelus</i>	Rhabditidae
<i>Ethmolaimus</i>	<i>Rhabdolaimus</i>
<i>Eucephalobus</i>	<i>Tobrilia imberbis</i>
<i>Eudorylaimus</i>	<i>Tobrilus nicasimilis</i>
<i>Filenchus</i>	Tylenchidae
Heteroderidae	<i>Tylencholaimellus</i>
<i>Hirschmanniella pomponiensis</i>	<i>Tylencholaimus</i>
<i>Labronema</i>	<i>Tylenchorhynchus</i>
<i>Labronemella czernowitziensis</i>	<i>Tylenchus</i>
<i>Mesodorylaimus</i>	<i>Tylocephalus</i>
	<i>Ypsylonellus</i>

The nematode communities found at SRPER constitute a mixture of species typical for grasslands and/or freshwater sediments. Surprisingly, many “aquatic” nematodes were still recovered in November, five months after the last significant rainfall and just a few days after some very light rain. These include large species (by nematode standards) like *Dorylaimus* and *Paractinolaimus*, which are not known to be tolerant of desiccation. Furthermore, their occurrence in samples of heavy clay would seem to contradict established correlations between average nematode body size and soil texture. This apparent contradiction was partly resolved by mist chamber extractions of the roots and plant debris collected in the November samples: most of these large nematodes appear to survive dry periods within or between this organic debris, rather than inside the soil itself.

Other interesting nematodes found include a species of *Hirschmanniella* and *Tobrilia*. The first was provisionally identified as *H. pomponiensis*, a species originally described from Pomponio State Beach (San Gregorio, CA). Nematodes in this genus are specialized

parasites of plants growing in wet to waterlogged soil. The genus *Tobriolia* is considered extremely rare, only two species are known and their original descriptions constitute the only published reports in the entire nematological literature to date.

As our extraction procedures were designed for small nematodes, the unexpected importance of roots and plant debris as a reservoir precludes accurate quantitative analysis with current methods as approved in our state and federal permits. We are therefore preparing a request for amendment to our procedures, to allow us to apply longer mist extractions to roots and larger plant debris. However, it is of concern to us that the US Fish and Wildlife Service's processing time for any permit-related matters exceeds eight months, even for minor matters such as adding the name of a new staff member. For the time being, we will continue our sampling and interpret the results only in qualitative respects.

#### ***Numbers of cysts extracted and returned to sampling site***

Our state and federal permits require us to collect all fairy shrimp cysts recovered from our samples and return them to the respective sampling sites. The numbers of cysts recovered from our samples in 2003 are:

sampling date	6/12/03				11/3/03			
sampling site	L1	L2	C1	C2	L1	L2	C1	C2
intact cysts	2	2	2	0	26	1	11	1
broken cysts	2	0	0	0	13	0	2	0

The total numbers of fairy shrimp cysts encountered are very small compared to the overall cyst bank at either pool, which probably contains billions of cysts. The large differences between successive samples from the same sites is probably due to a combination of their patchy distribution and our increasing skills at recognizing them among other particles. Our methods do not allow us to distinguish between breakage caused by our processing, versus breakage due to natural causes (i.e. prior to sampling), but even in a worst case scenario we estimate to have caused mortality no greater than one third of the extracted cysts.

No obvious evidence of cyst puncturing or parasitism by nematodes was encountered so far, but based on our current experience we suspect different observation tools are required to discover such evidence. All cysts found in these two sampling series belonged to the genus *Branchinecta*, so presumably they are cysts of the vernal pool fairy shrimp (*B. lynchi*).

#### ***Preliminary conclusions***

No nematodes were encountered from groups that are indicative of eutrophication, bacterial blooms or toxic pollutants. This suggests that the pools have so far not suffered from significant pollution that could be caused by e.g. runoff from nearby residential or agricultural areas. Some of the encountered large nematodes are known to be voracious predators, and it is possible that their diet includes fairy shrimp cysts. Biogeographic

records for these and related nematodes are very scarce in California, but it is quite likely that they are widespread and a natural component of freshwater ecosystems.

Among the plant parasites, *Hirschmanniella pomponiensis* is unlikely to represent a threat to any of the endangered plants, as this species may well be a true native of the Santa Rosa Plateau. The occurrence of the family Heteroderidae requires further study, as this family includes many of the most important economically damaging plant parasites. Fortunately, this also means that diagnostic DNA sequences are available for a rapidly increasing number of species in this family, and molecular identification is in progress of this and other species from SRPER.

### **Student support**

	Total Project Funding		Supplemental Awards	Total
	Federal Funding	State Funding		
Undergrad.	2	2		2
Masters				
PhD.				
Post-Doc.			1	1
Total	2	2	1	3

Note: two undergraduate students (Mrs Sheila Esfahani and Mrs Melissa Yoder) were directly supported by project funding, each partly from state and partly from federal funding. Hence the total of 2 for the Undergraduate section above. Funding from other sources (UCR startup funds) was used to support project-related activities by one Post-Doctoral Researcher (Mrs. Irma Tandingan De Ley).