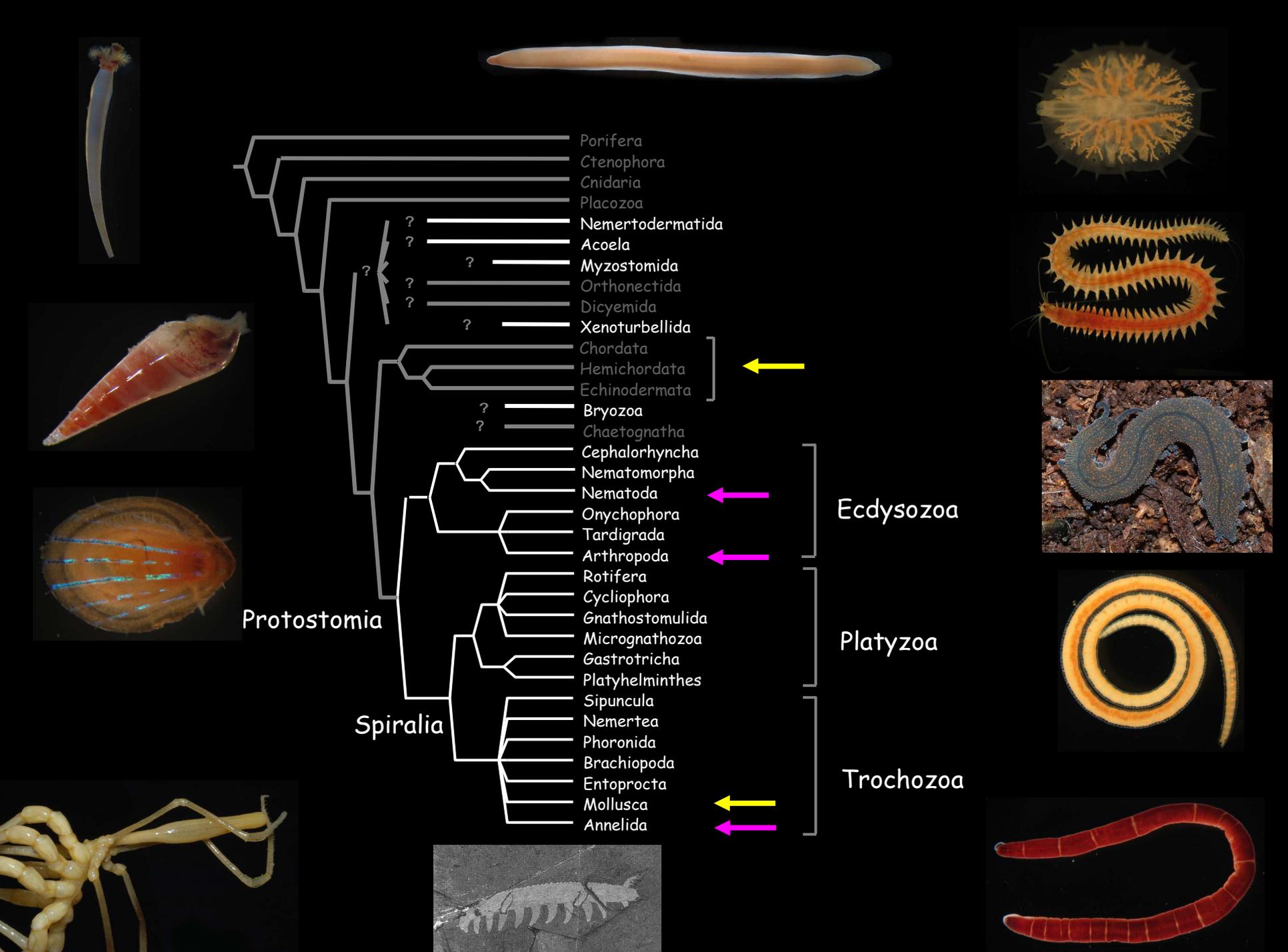


An integrated approach to the origin and diversification of protostomes

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<http://www.mcz.harvard.edu/Departments/InvertZoo/tol/>



Project Goals (2 years):

1. Generate a **collection of tissues for 265 species** belonging to all protostome phyla. Having such a collection available for research will ensure gathering of different sources of data for the same organisms across laboratories worldwide.
2. Generate sequence data (**about 10 markers** that have been shown to resolve metazoan deep divergences) for 50% of the metazoan lineages proposed in the previous section.
3. Generate **new cDNA libraries for 8 protostomes**.
4. Generate sequence data for **about 100 molecular markers by EST technology** for the ca. 15 taxa for which the cDNA libraries are generated/available.
5. Generate **new embryological cell lineage data** for a selected collection of protostome phyla for which cDNA libraries have been generated.
6. Generate **gene expression data** for several genes implied in segmentation, mesoderm, and body plan organization.
7. Compile a **comprehensive data set on protostomes morphology/anatomy** coded for **exemplar taxa**, including key fossil species (see accessory Table).

Goal 1:

- Fieldwork has been conducted in Bermuda, Florida, France, Japan, Australia and Sweden
- Tissues of more than 200 species belonging to Acoela, Xenoturbellida, Myzostomida, Nemertea, Mollusca, Annelida, Sipuncula, Entoprocta, Brachiopoda, Phoronida, Platyhelminthes, Gnathostomulida, Gastrotricha, Rotifera, Cyclophora, Micrognathozoa, Priapula, Kinorhyncha, Tardigrada, Loricifera, Onychophora, Arthropoda have been collected and preserved in RNAlater, 96% EtOH, formalin, and glutaraldehyde
- Online database of species, modes of preservation, and status of work is under development. A preliminary version can be viewed at:



ATOL Checklist

<http://collections.oeb.harvard.edu/Invertebrate/atol/spi>

Haplognathiidae

- Haplognathia gubbarnorum* (Sterrer, 1969)
- Haplognathia lunulifera* (Sterrer, 1969)
- Haplognathia rosea* (Sterrer, 1969)
- Haplognathia ruberrima* (Sterrer, 1966)
- Haplognathia simplex* (Sterrer, 1966)

Pterognathiidae

- Cosmognathia aquila* Sterrer, 1998
- Cosmognathia arcus* Sterrer, 1991

Kinorhyncha

Echinoderidae

- Echinoderes horni* Higgins, 1983
- Echinoderes n.*

Pychnophyidae

- Pychnophyes groenlandicus* Kristensen & Higgins, 1984
- Pychnophyes sp.*

Mollusca

ATOL Species Card

<http://collections.oeb.harvard.edu/Invertebrate/atol/specie>

browse records

Record Number	
Phylum	Class
Xenoturbellida	
Order	Suborder
Family	
Xenoturbellidae	
Taxonomic Name (Genus species)	Author, year
Xenoturbella bocki	
Depository 1	Depository 2
Depository 3	Depository 4
Depository 5	
Locality:	Sweden: Tjärno
Coordinates:	
Collector	Date
AToL expedition 2004	July 2004
Habitat:	Muddy bottoms
Preservation	Data
<input type="checkbox"/> live specimens	<input checked="" type="checkbox"/> CDNA Library
<input checked="" type="checkbox"/> RNA Layer	<input checked="" type="checkbox"/> ESTs
<input checked="" type="checkbox"/> 96% Ethanol	<input checked="" type="checkbox"/> RNA
<input checked="" type="checkbox"/> Glutaraldehyde	<input checked="" type="checkbox"/> DNA
<input checked="" type="checkbox"/> Paraformaldehyde	<input checked="" type="checkbox"/> Histology
	<input checked="" type="checkbox"/> TEM

Record 117 of 117

[<- Previous Record](#)

<http://collections.oeb.harvard.edu/Invertebrate/atol/species.cfm>

Goals 3 and 4:

cDNA libraries have been built for 5 protostomes:

Myzostoma seymourcollegensis n. sp. (Myzostomida)
Cerebratulus lacteus (Nemertea)
Themiste lageniformis (Sipuncula)
Xenoturbella bocki (Xenoturbellida)
Phoronis vancouverensis (Phoronida)

ESTs have been sequenced for 5 animals:

Mnemiopsis leidyi (Ctenophora), 1,000 clones
Capitella sp. I (Annelida), 1,000 clones
Myzostoma seymourcollegensis n. sp. (Myzostomida), 1,400
Cerebratulus lacteus (Nemertea), 900 clones
Themiste lageniformis (Sipuncula), 100 clones (in progress)
Xenoturbella bocki (Xenoturbellida), 3,000 clones (in progress)

We have obtained homologues of several markers that could be useful in phylogenetics such as tropomyosin, elongation factors, α -ubulin, Dynein, Notch, myosins, etc.

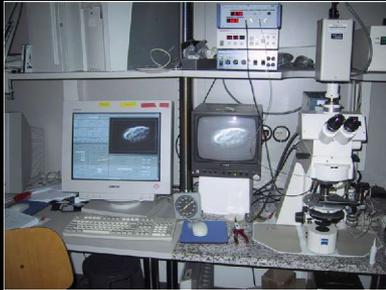
Brief protocol: Specimens are collected in the field, preserved in RNAlater, transferred to Kewalo for generation of cDNA, plates are sent to the sequencing facilities of the AMNH or Harvard.

Next protostomes: acoels, gnathostomulids, priapulans, aplousobranchs, pycnogonids...

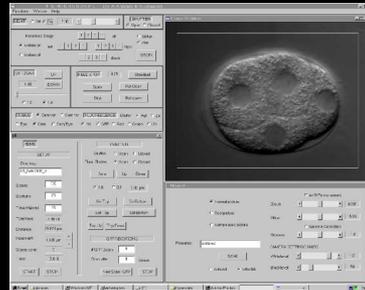


Goal 5:

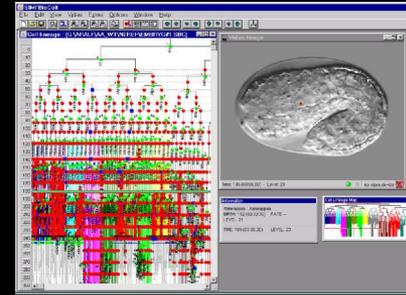
Cell lineages for selected species of metazoans. An example from the gastrotrich *Lepidodermella squamata*. A new technique using a 4D microscope has been developed to study cell lineages in small invertebrates. With this technique we are able to expand the number of embryos that can be studied for cell lineages without the hassle of cell injections.



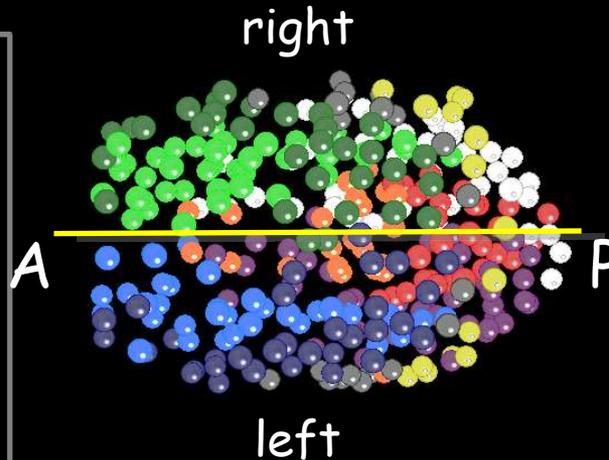
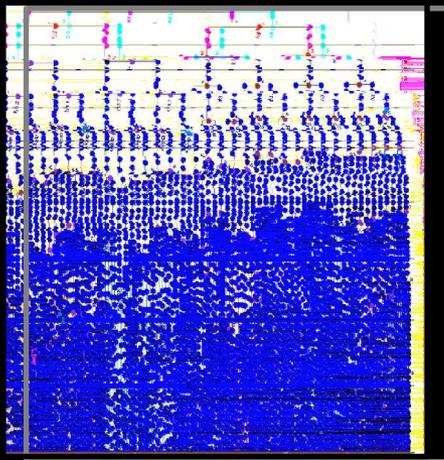
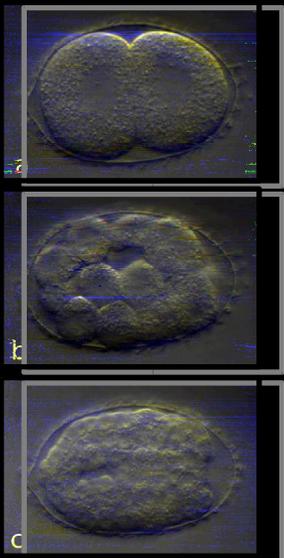
Motorized microscope



Trigger software



Analysis software



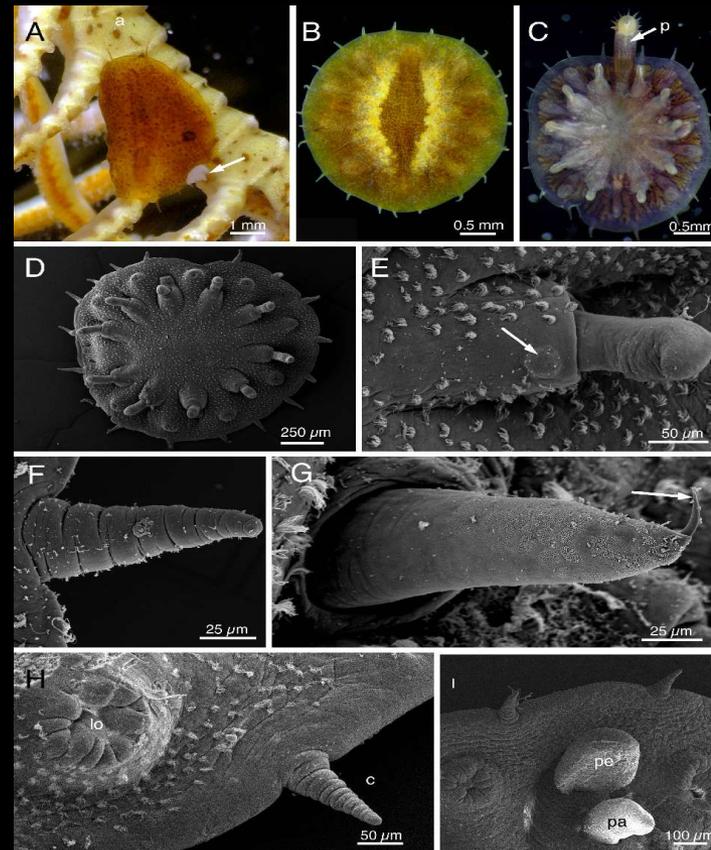
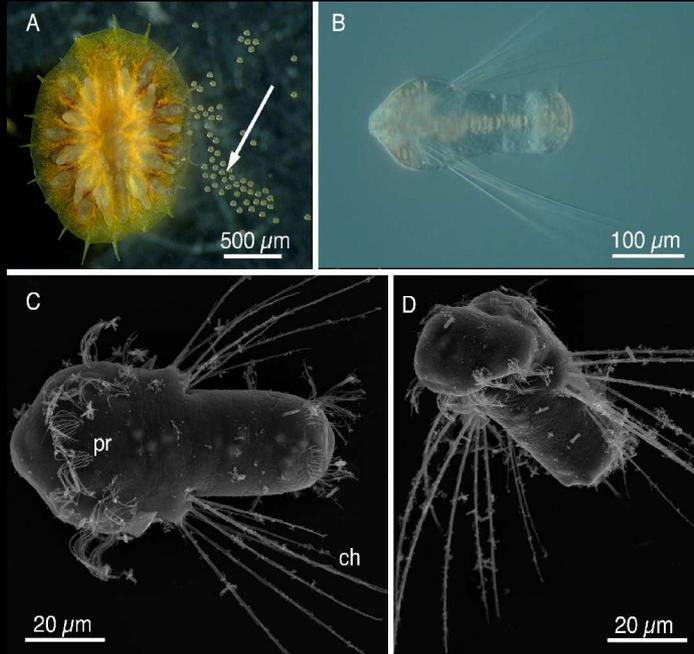
-  dorsal ectoderm left
-  mesoderm left
-  mesoderm right
-  dorsal ectoderm right
-  ventral ectoderm right
-  neuronal ectoderm
-  ventral ectoderm left
-  pharynx
-  gut

Cell Fates

Case Study: An unknown protostome species is discovered and studied; The "quasi-model" approach

- Description and morpho-anatomical characterization of animal (light microscopy, SEM, TEM)
- Spawning for study of larvae and developmental stages
- Generation of cDNA library
- Sequencing of ca. 1,400 ESTs
- Study of expression patterns

We will apply this integrated approach to a number of key species to understand protostome evolution, including *Xenoturbella*, acoels, and others.



This test study was designed to demonstrate that we can develop almost any invertebrate into a "quasi-model"