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**Larval Development of the polychaete annelid, *Serpula vermicularis***

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## Introduction

*Serpula vermicularis* is a tube-dwelling, filter-feeding worm. It is found in oceans worldwide, and has even been found as *Serpula vermicularis* reefs in Scotland (1). *Serpula vermicularis* is dioecious, with separate male and female sexes (2). It is a polychaete annelid, in the family Serpulidae (3). These organisms generally develop into trochophore larvae.

Trochophore larvae have a characteristic mode of feeding, where small particles are captured between two ciliary bands, and carried to the mouth by a food groove (4). This mode of feeding can be significantly impaired by lowering temperature, due to increased water viscosity at lower temperatures, as well as physiological effects of lower temperature (5). As trochophore larvae metamorphose, they gradually lose larval structures and acquire adult structures including jaws and chaetae (3).

Once the trochophore larvae becomes a juvenile, it will eventually choose a site to settle. There are a number of possible settlement cues that have been identified for serpulid worms, including light, chemicals, depth, and hard surfaces (6-9). Serpulid worms, including *Serpula vermicularis* and *Pomatoceros triqueter*, preferentially recruited to the underside of horizontal panels (8). They also preferentially recruited to panels at 7 meters compared to panels at 4 meters (8). This preference could be mediated by responses to light, as *Serpula vermicularis* shows an interesting reaction to light that changes over the course of development. When settling, *Serpula vermicularis* orient their primary tube aperture away from light (6). Finally, there is evidence for chemical settlement cues in serpulid worms, as evidenced by the relationship between the polychaete *Hydroides elegans* and the bryozoan *Bugula neritina* (7).

In order to observe these developmental stages, I started a culture of *Serpula vermicularis*. I raised the culture in a cold, dark environment (4-10° C). Based on previous work, these conditions should retard feeding and therefore development of the *Serpula vermicularis*. However, the trochophore larva and the juvenile feed differently, as the juvenile may have more adult feeding structures. The increased viscosity of the cold water should hinder feeding by the trochophore larva, but the effect of cold temperature on the juvenile may be different. To begin to look at these questions, I observed the development of *Serpula vermicularis*.

## **Materials and Methods**

Adult *Serpula vermicularis* were collected on May 1, 2007, by trawling near the coast of Charleston, Oregon. Additional *Serpula vermicularis* were harvested from boat docks in Charleston, Oregon. To induce release of gametes, the *Serpula vermicularis* tubes were scraped off the rocks they were settled on. This induced some of the adults to release gametes. Sperm and eggs from several different individuals were mixed together in seawater to begin the *S. vermicularis* culture.

Cultures were maintained in filtered seawater, and larvae were fed with *Dunaliella teriolecta* and *Rhodomonas lens*. Filtered seawater for the culture was changed every two to three days, and algae was added to the culture at the same time. Cultures were maintained in the dark at 4-10° C for 10 days. At this point, incubator failure led to the collapse of the culture. Cultures were re-started from a stock culture that had been maintained at the ambient seawater temperatures of Charleston, Oregon. Data from day 16 forward is from these re-started cultures. These new cultures were

maintained at 4-10° C for the remainder of the observation, with one anomalous temperature spike to 30° C. Despite this temperature spike, the cultures continued to be survive. Therefore, observation was continued for a total of 34 days.

## Results

*Serpula vermicularis* shows a pattern of spiral cleavage, which can be discerned from the embryo at the 8-cell stage. By day 2, *Serpula vermicularis* is a blastula. By day 5, the prototroch of *Serpula vermicularis* has developed. By day 7, the prototroch and metatroch can be seen on the larva. Three days later, the stomach is clearly visible, and the other structures of the trochophore larvae easily discerned. This larvae has a long apical tuft, a prototroch and metatroch that surround a food groove, and an anal vesicle.

On day 16, the *Serpula vermicularis* larvae have developed chaetae and left and right ocellus. Further development shows an increase in segmentation, with the larvae's chaetigerous segments becoming more defined. On day 23, the larvae continues to elongate. By day 28, the larva's head has changed shape, such that it is more separate from the body. The left and right ocellus have moved closer together. At this point, the larvae have radiole buds, which will later develop into adult feeding structures. The gut of the animal has moved further down into the body. However, by day 34, the larvae in this culture no longer look healthy.

These observations were used to make an approximate developmental timetable, showing when major structures developed over time (Table One). Observations were based off a small number of larvae, so there is likely to be more variation in development time than I have sampled in this study.

**Table One:**

	prototroch	metatroch	Apical tuft	stomach	chaetae	ocellus	Antenna	Radiole bud
Day 1								
Day 2								
Day 5	X							
Day 7	X	X						
Day 10	X	X	X	X				
Day 16	X	X		X	X	X		
Day 17	X	X		X	X	X		
Day 19	X	X		X	X	X		
Day 21				X	X	X		
Day 23				X	X	X	X	
Day 28				X		X		X
Day 34				X				X

## Discussion

The development of my first culture of *Serpula vermicularis*—from fertilized egg to trochophore larva—took considerably longer than was seen in previous studies. At 4 degrees Celsius, the trochophore larva was first seen at seven days. Previous studies observed trochophores at three days (6). The second culture of *Serpula vermicularis* did not show retarded development. This may be due to the difference in feeding structures between juveniles and trochophores, as the increased viscosity of cold water may be more detrimental to trochophores.

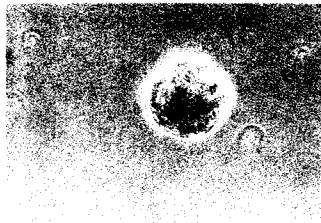
According to Charles Zeleny, the attachment phase of the serpulid larva is the stage that is most likely to lead to death. None of the *Serpula vermicularis* culture settled during my study. Settlement of *Serpula vermicularis* takes 40-50 days, so if I had continued my study for longer, There are many possible settlement cues for serpulid worms, including light, chemicals, depth, and hard surfaces (6-8). In particular, it has been shown that a response to light is important in the ontogeny of *Serpula vermicularis*. It is possible that the lack of light in my study hindered the ability of *Serpula vermicularis* to effectively settle, or perhaps another cue was necessary for settlement.

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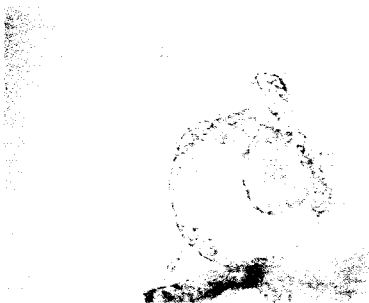
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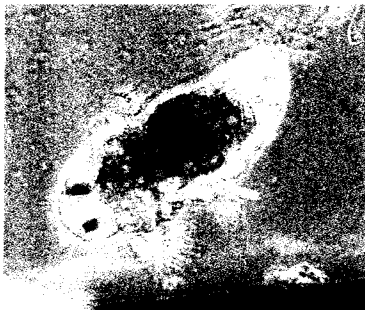
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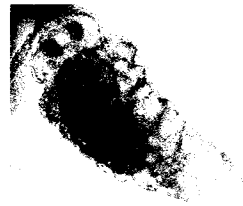
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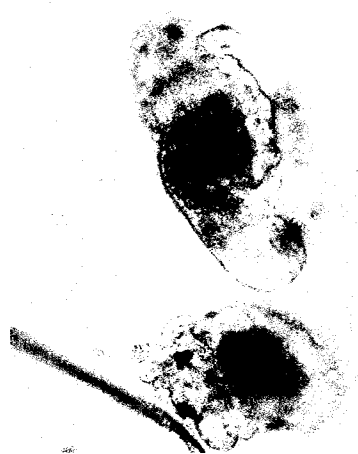
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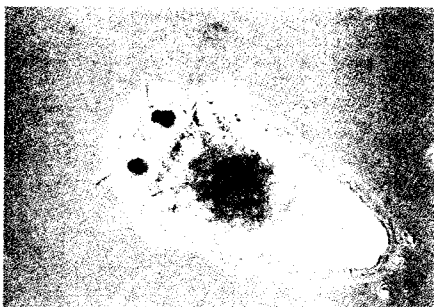
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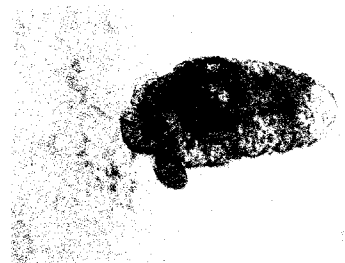
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