Keynote Presentation

Wednesday Morning September 5rd

Weapons of Micro-Destruction: An Interdisciplinary Approach to Understanding a Parasitic Cnidarian

Jerri L. Bartholomew

Department of Microbiology, 226 Nash Hall, Oregon State University, Corvallis, OR 97331 USA Jerri.bartholomew@oregonstate.edu

Myxozoa are now understood to be an ancient, diverse group of endoparasitic Cnidaria. On their route to parasitism, myxozoans have become morphologically and genetically simple, losing many traits of their free-living relatives. While most cnidarians have life cycles that alternate between a medusa and a polyp, myxozoans alternate between two different spore stages (e.g. myxospore and actinospore) that develop in two different hosts (e.g. fish and aquatic annelid. Myxozoans have retained the phylum-defining stinging organelles known as cnidae or polar capsules, but we are only just beginning to understand what features have been lost or modified to suit their parasitic needs.

Free-living Cnidaria like corals, jellyfish and anemones, use their stinging capsules for prey capture or defense, but in Myxozoa these capsules have the essential function of initiating the host infection process. To better understand the evolutionary relationships between myxozoan polar capsules and cnidae in free-living species, we examined the structure and firing behavior of capsules in 4 myxozoans, including *Myxobolus cerebralis*, the causative agent of whirling disease in trout, and *Ceratonova shasta*, the cause of enteronecrosis in salmonids. Electron microscopy and high speed video analyses showed that the three *Myxobolus* species had highly elastic polar tubules, which is a property unknown in free-living cnidarians. By dye-labeling the polar capsules prior to firing, we discovered that two of the species could release their entire capsule content, which suggested that cytotoxic or proteolytic compounds may be present in the capsule, and are injected into the host to facilitate infection. Moreover, while free-living cnidarians inject toxins through the tip of the tubule, we identified pores along parts of tubules of *Myxobolus* species, and showed that the tip of the tubules was sealed. In contrast, *C. shasta* and *M. cerebralis* tubules had no openings at all and no apparent delivery of capsule content, thus are likely used simply to anchor the spore to the host without any toxin injection.

To examine functions of polar capsules at a proteomic level, we isolated *C. shasta* capsules using a dielectrophoresis-based microfluidic chip platform. In comparison to jellyfish, sea anemone and *Hydra*, which share 49 protein domains, only 27 domains were shared between these Cnidaria and *C. shasta*. While *C. shasta* capsules have retained typical structural and housekeeping proteins found in free-living Cnidaria, they have lost toxin-like proteins, which supports the structural evidence of the video analysis. Our findings indicate that although polar capsules and nematocysts are homologous organelles, myxozoan capsules have more functions

than previously assumed. Understanding these mechanisms may provide a means to prevent myxozoan infection to fish by interfering with the initial stages of the infection process.

The ultimate function of myxozoan cnidae is to sense and respond to proximity with the host, then fire the tubule within a fraction of a second, to anchor the spore to the host. This process initiates the infection process, but must be followed by invasion of the parasite sporoplasm into the host tissues, which requires motility. We used *C. shasta* as a model to study motility because we have a reference transcriptome and different parasite genotypes with varying degrees of virulence. Developmental stages of *C. shasta* showed a variety of locomotion mechanisms, using amoeboid movement, filopodia, lamellipodia and blebbing to migrate between host cells. We mined the *C. shasta* reference transcriptome for motility genes that may serve as virulence factors. Here we looked at genes that may be involved in the actomyosin machinery of the cell, in cell adhesion, and in cell motility regulation. The more virulent genotype had increased levels of adhesion factors that connect the parasite cytoskeleton with the host extracellular matrix, which suggests that these components are virulence factors of the parasite. In addition, examination of the transcriptome for proteases shows that proteases were expressed in the developmental stages and these may also function as virulence factors, facilitating parasite invasion.

Our changing vision of what myxozoans are has been informed by engaging researchers in other scientific fields, but we can also learn something by stepping outside our scientific silos to gain perspectives and inspiration from other disciplines. To explore how the humanities contributes, or enhances, our understanding and appreciation of our science, I will weave in my own artistic interpretations as well as collaborations with poets, artists and musicians.