Discover the Microbes Within!: The *Wolbachia* Project

Seth R. Bordenstein
The Marine Biological Laboratory,
Woods Hole, Massachusetts

Seth Bordenstein received a B.S. in ecology and evolutionary biology, an M.S. in biology, and a Ph.D. in evolutionary genetics from The University of Rochester. He was a National Research Council Postdoctoral Fellow at the Marine Biological Laboratory (MBL). In 2005, Dr. Bordenstein joined the faculty of the MBL's Josephine Bay Paul Center as an assistant scientist where he studies the mechanism and relevance of bacterial endosymbiosis. He is also a faculty member in the Department of Ecology and Evolutionary Biology at Brown University and a founding member of the Mobile Genetic Element Cluster at the MBL.

Project website: http://jbpc.mbl.edu/~sbordenstein/workshop.html

Project description: The *Wolbachia* Project is an integrative lab series and professional development workshop on biotechnology, real-world research, and microbial symbiosis.

Discover the Microbes Within!: The *Wolbachia* Project is designed for high school and undergraduate educators in an effort to modernize biology labs and lesson plans with inquiry, discovery, biotechnology, and microbial symbiosis. The project is sponsored until 2012 with gracious support from the Howard Hughes Medical Institute's Precollege Science Education Program. The project's four broad goals are to: (i) engage high school and undergraduate students in inquiry and real-world research, (ii) encourage nationwide participation in the collection and reporting of new scientific data on bacterial endosymbionts, (iii) enhance student interest and understanding in biology through an integrated lab series spanning biodiversity to molecular biology, and (iv) show students what it is like to be a scientist. The primary objective is to challenge students in real-world research throughout the calendar year with laboratory exercises and summer research experiences on *Wolbachia*, a widespread bacterium that lives symbiotically in at least 20 percent of the world's insects. This objective is founded on the principle that students naturally want to learn science the way science is done, and students who conduct their own science will enhance their understanding of scientific inquiry while also collecting reliable data. Discover the Microbes Within! excels in this aspect as it teaches observation, conceptualization, the scientific method, and major concepts in systematics and biodiversity, genomic DNA isolation, biotechnology, DNA sequencing, bioinformatics, and molecular evolution. These six lab modules are designed to be either individually incorporated into daily lesson plans addressing National Science Education Standards or used as a coherent unit progressively emphasizing the nature of a long-term science project throughout the school year.

Initiated in 2005 at a Marine Biological Laboratory (MBL) professional development workshop with support from the National Science Foundation and NASA Astrobiology Institute, the lab series moves seamlessly from studying organisms to molecules and begins with students discovering the spectacular diversity of insects through insect collections, photo vouchering, and a web-based tool that modernizes insect classification. This first unit establishes student ownership of the experiments as students decide which insects to collect and study, rather than being given an insect and told what to do in a predetermined way. The students will find themselves asking, just as scientists do, "Does my insect have *Wolbachia* endosymbionts within it?" and "How can I determine this?" Students will then gain an understanding of DNA as the genetic material that connects all life and the forensic tool to investigate the vast unseen world of symbiotic bacteria in insects. In doing so, students will use biotechnology techniques including DNA extraction, polymerase chain reaction, and gel electrophoresis to investigate whether local insects contain the bacterial endosymbiont *Wolbachia*. We expect that a student will diagnose *Wolbachia* in one out of five insect species that (s)he collects based on worldwide sampling estimates. The symbiont survey thus yields an end product (i.e., infection status of classified insects) that will be useful to the scientific community, as numerous insects will surely be discovered to harbor *Wolbachia* that were unknown to the scientific community.

For those insects that test PCR positive for the 16S rDNA gene of *Wolbachia*, students will confirm their findings and then partner with the Bordenstein lab for sequencing of *Wolbachia* 16S rDNA. Classes will ship PCR products to the MBL and a technician will produce DNA sequences that are deposited in a website repository. Students will learn to use the National Center for Biotechnology Information (NCBI), to study how professional biologists organize and analyze genetic sequence information. An online bioinformatics unit will teach students to use the various taxonomy functions of NCBI and simple BLAST exercises. Such computational tools are universal to biologists today and immediately foster student inquiry in a free and accessible way. The student sequence data will be freely retrievable from the MBL onto classroom and home computers. One of the most exciting aspects is that it will provide students with a forum to publish their DNA sequence data. The *Wolbachia* DNA sequence is another end product of the innovative lab series. It is an automatic motivator because it is publishable and accessible by students, teachers, families, and scientists. Upon retrieval of DNA sequences, students will BLAST for sequences similar, different, or identical to their own *Wolbachia* DNA in the published sequence database. In

the last laboratory, students will construct a DNA-based phylogenetic tree with the sequences that they generate. Students will study genetic relatedness, evolutionary relationships, and classification methods based on DNA sequence information.

Finally, the program will offer summer "envisionships" or research experiences for 6 weeks in a MBL or alternate university lab for students who participated in the project during the school year. Partnerships with leading *Wolbachia* scientists will contribute to a nationwide top-down model where *Wolbachia* biologists serve as summer hosts in their laboratories. The director will select students based on an essay about his/her experience, a teacher recommendation letter, and the availability of a regional scientist interested in developing the envisionships. The envisionships aim to break down perceived disconnects between students and scientists and to crystallize in students' minds what it is like to be a scientist. They are also effective in enhancing student skills and critical thinking, and influencing some participants to further consider the life sciences in college.

In addition to the hands-on training, Discover the Microbes Within! will assist teachers in using the curriculum in their classrooms by connecting them with scientists in the field and with teacher emeriti of the project. The program will also provide travel assistance for teacher meetings and offer Internet forums with other teachers who wish to get involved. Discover the Microbes Within! is in a significant growth phase and seeks new teachers to implement the lab series. Please visit the website above for downloads and full information on protocols, lectures, annual workshop, and general information.

Over the past 3 years, this discovery-based lab series engaged 90 teachers and 2,000 students in real-world research on molecular biology, evolutionary biology, and biotechnology. Regional centers and teachers that are pioneering this modern lab series on *Wolbachia* symbionts include Falmouth Public Schools (Falmouth, MA; Christine Brothers), Fannie Lou Hamer Freedom High School (Bronx, NY; Molly Shabica), and Loudon County Schools (Loudon county, VA; George Wolfe and Jackie Curley). Faculty from the MBL workshop include Seth Bordenstein (MBL), Michele Bahr (MBL), Jack Werren (University of Rochester), Laura Baldo (University of California), Michael Clark (University of Rochester), Bob Minckley (University of Rochester), Jennifer Wernegreen (MBL), and George Wolfe (Loudoun Academy of Science).

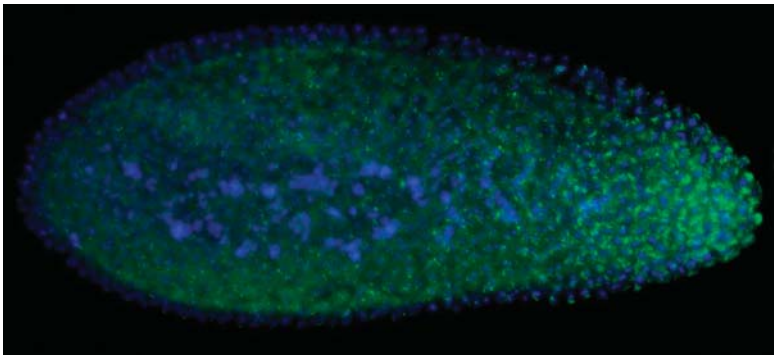

FIG. 1. *Wolbachia* in an insect egg. A wasp egg showing DAPI-stained insect nuclei (blue) and maternally-transmitted *Wolbachia* endosymbionts (green) localized towards the posterior tip of the embryo. *Wolbachia* are intimately associated with millions of arthropod species and can profoundly influence the ecology, evolution, reproduction, and development of their host insects. Photo copyright Seth Bordenstein and Michael Clark, 2007.