PROJECT SUMMARY.

We propose to assemble a comprehensive collection of full-length cDNAs representing the preponderance of genes expressed during the life cycle of the parasitic ciliate *Ichthyophthirius multifiliis*. *Ichthyophthirius* is a major pathogen of freshwater fish and has substantial impact on aquaculture in this country and abroad. From a biological standpoint, it is one of the few obligate parasites within the ciliate taxon, and has a variety of unique features related to its cell cycle, and interactions with the host. Because *I. multifiliis* has a relatively large genome (~ 100 Mb) that is highly AT-rich (> 75%), we believe that high-throughput sequencing of expressed genes represents the best approach towards unraveling the coding potential of its genome under the scope of the Microbial Genome Sequencing Project. Libraries will be prepared from mRNAs derived from parasites at different stages of the life cycle, and under conditions of immune stress. Overall, the intent of our proposal goes beyond bioinformatics, and extends to the development of an archival resource that can be used by the wider community in the design and manufacture of potential vaccines and other therapies for the prevention and treatment of "white spot". The project will be undertaken entirely within an academic setting and has been expressly designed to include the training of undergraduate and graduate students in a range of disciplines that span bioinformatics, computational biology and molecular genetics. This effort at training and outreach is considered one of the most important aspects of the proposal. To our knowledge, no similar efforts are being carried out with *I. multifiliis* or any other parasitic ciliate. Over the past 50 years, the ciliates have contributed enormously to our understanding of cellular and molecular biology. Despite this, a paucity of genomic sequence data exists for the phylum as a whole. High-throughput sequencing efforts are currently slated for 3 free-living species (namely, *Tetrahymena thermophila*, *Paramecium tetraurelia* and *Oxytricha trifallax*), and the work proposed here should lend to a more complete understanding of ciliate biology and evolution. At the same time, we hope to gain fundamental insights into the nature of virulence and parasitism in *I. multifiliis*.