Understanding the role of parasites in salmon mortality

The decline of Pacific salmon has had its full share of controversy and conflict. Certainly one of the most controversial incidents in recent years was the die-off of an estimated 34,000 fish in the Klamath River in September 2002. Although this loss was dramatic because it involved adult fish, the epidemic drew attention to the chronic disease issues that affect the health of migrating juvenile fish. What should be done to improve the health of these fish and ensure that such incidents don't recur is a question with consequences for the farmers, fishers, and communities that depend on the river's water in Oregon and California, as well as for the tribes and agencies that manage the river.

Lynn Ketchum, OSU Extension and Experiment Station Communications

Jerri Bartholomew, who says that *C. shasta* infections have been detected in about 35 percent of juvenile Chinook salmon—about half of which will die.

Fish disease issues in the Klamath River are complicated. The U.S. Fish and Wildlife Service (USFWS), in its report on the 2002 fish die-off, concluded that a "combination of factors" caused it, specifically, "high density of fish, low [river] discharges, warm water temperatures, and possible extended residence time of salmon [which] created optimal conditions for parasite proliferation." An epidemic outbreak of two pathogens, Ich (*Ichthyophthirius multifiliis*) and columnaris (*Flavobacterium columnare*), was the "proximate cause of death," the agency said. However, fish health-monitoring studies conducted by the USFWS show that mortality in juvenile salmon results primarily from infections caused by myxozoan parasites. As many as 45 percent of juvenile Chinook salmon captured in 2004 in the lower Klamath River were infected with one particularly injurious species, *Ceratomyxa shasta*, which infects the intestine. Prevalence of infection by another myxozoan, *Parvicapsula minibicornis*, which infects the kidney, can be greater than 90 percent. Infection can reach 100 percent for both parasites in fish migrating during spring.

As agencies and the courts have become involved in attempting to balance the needs of the river's human users, fish, and other wildlife, a key concern is limiting fish disease caused by parasites. Research on these pathogens is a specialty of an Oregon State University (OSU) scientist, Jerri Bartholomew. Bartholomew, a microbiologist with the OSU Center for Fish Disease Research, has investigated the life cycles of these parasites, their distribution in the Pacific Northwest, and their effects on salmon. Additionally, her laboratory has developed molecular methods to better enable detection of the parasite.

Breakthroughs in parasite detection

Until now, researchers had no quick, easy way to test for parasite abundance. The assay developed by Bartholomew and co-worker Sascha Hallett with funding from Oregon Sea Grant uses the organism's own DNA. The method, known as quantitative polymerase chain reaction (QPCR), can detect even 1/1000th of a parasite spore in a water sample.

In the QPCR, a small sample of DNA is copied multiple times so it can be used for analysis, such as is done in genetic fingerprinting and paternity testing. A fluorescent tag is used to track the reaction so that the amount of accumulated PCR product can be measured.

To detect *C. shasta* before this breakthrough, scientists had to maintain fish in cages along areas of the river suspected to be infectious and then return them to the

laboratory and wait for months to see if clinical signs appeared. There was no way to quantify the number of infectious spores moving through the water.

Bartholomew and her partners used the QPCR methodology to investigate the distribution of *C. shasta* in the Klamath River. The parasite was detected throughout the river, and two of five tributaries tested contributed parasites to the main stem. Several sites were found to have parasite abundance in excess of 20 spores per liter.

"This is a huge advance in what we're able to do," said Bartholomew. "We wanted to offer a tool that would be useful if managers were to test management options like altering flows at certain times of year, so that effects could be determined immediately."

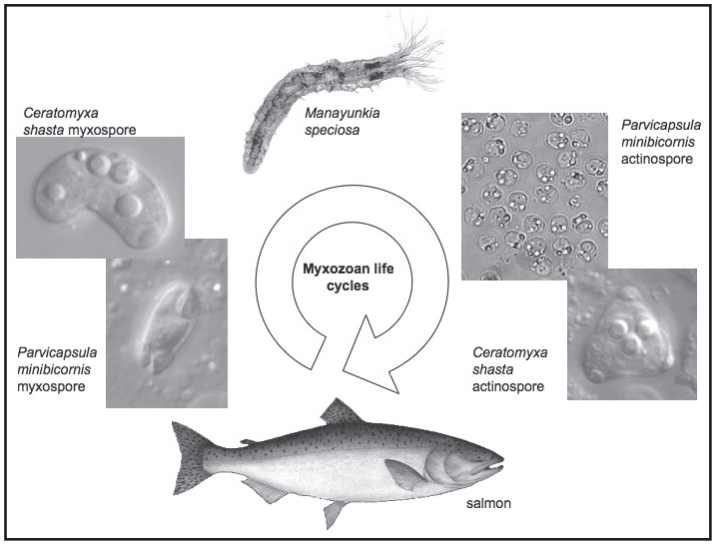
Disease epidemiology

Another significant finding of Bartholomew's Sea Grant-supported work is that there is a distinct difference in the severity of *C. shasta* infection in fish exposed in different portions of the Klamath River. Specifically, in the reaches below Iron Gate Dam, Chinook salmon are exposed to high parasite numbers that are responsible for the high mortality documented in outmigrating juvenile fish. Above the projects, infection patterns are variable, and particularly in reservoirs, exposure is low and not likely to result in mortality. The life cycle of *C. shasta* is established in the main stem of the Klamath, with little contribution from the tributaries.

Parasite life cycles

C. shasta was previously discovered to have a two-host life cycle with development in both a fish and a polychaete worm (*Manayunkia speciosa*) (see illustration). One additional result of Bartholomew's Sea Grant project has been the discovery that the life cycle of *P. minibicornis* is very similar, requiring the same worm host. This organism was only recently identified in the Klamath system. However, the parasite is considered a contributor to mortality of adult salmon in rivers in British Columbia and has been detected in the Columbia River Basin. The finding that these parasites share the same host will have important implications for management of these diseases.

During the past two years, a graduate student in the Bartholomew lab has conducted an extensive survey for polychaetes throughout the Klamath River. As a result, says Bartholomew, "we are beginning to understand why disease is so severe in certain locations." Polychaete distribution is highly patchy and influenced by in-stream primary productivity, flow, substrate embeddedness, and the presence of compact algal epiphytes such as *Cladophora* species. Infection rates in these polychaete populations also differ, contributing to variations in disease severity in fish.



Life cycle of *C. shasta* and *P. minibicornis*, showing release of actinospore stages of both parasites from the polychaete, infection of the salmon, and release of myxospore stages that infect the polychaete.

In a new research project with Oregon Sea Grant, Bartholomew focuses on both *C. shasta* and *P. minibicornis*. The objectives include the development of similar diagnostic methods for *P. minibicornis*, a comparison of the distribution and seasonal occurrence of these parasites in the Klamath River, a determination of the role of various salmonids in supporting the life cycle of these parasites, and a determination of the effects of the parasites on seawater survival of salmonids. "One of our goals is to provide information to those involved in salmon recovery that will enable them to more effectively manage the system to decrease the effects of the disease."

Collaborators with the OSU research team include the California-Nevada Fish Health Center; the U.S. Fish and Wildlife Service, Arcata, California; the Yurok, Hoopa, and Karuk tribes; PacifiCorp; and Humbolt State University.

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