CERATOMYXA SHASTA 2007 STUDY SUMMARY June 5, 2008 Investigator: Jerri Bartholomew, Department of Microbiology, Oregon State University Funding: Bureau of Reclamation, Oregon Sea Grant



SENTINEL FISH EXPOSURES:

Sentinel exposures are being conducted to determine:

- 1. How infection levels this year compare with levels in previous years.
- 2. How temperature affects disease rate.
- 3. If the distribution of the parasite has changed.
- 4. The relative susceptibility of Chinook and coho salmon
- 5. The relationship between parasite numbers measured in water samples and biological effects in the different fish species.

Exposures were conducted May 15-18, June 18-21 and Sept 22-14, 2007 for ~72 hrs at locations in the upper and lower river (**Figure 1**). Rainbow trout and Chinook salmon (IGH stock) were held at all sites; coho salmon (IGH stock) were held only at the site above Beaver Creek. In June, Trinity River steelhead were also held at the Beaver Creek site. Fish were held at average Klamath River temperature as well as at the normal laboratory water temperature of 13° C used during previous years. In May ambient water temperatures averaged 18° C, 20° C in June, and 18° C in Sept.

Rainbow trout (*C. shasta*-susceptible strain) were held at all sites at both temperatures to provide a baseline for between-year comparison. Similar to previous years, the general pattern of mortality in rainbow trout during May shows the Williamson River, Beaver Creek and Seiad Valley exposure groups having similarly high mortality (**Figure 2**). Mortality in the group held at Orleans was also high, but the mean time to death was decreased, indicating a lower exposure dose. As in previous years, mortality was low among fish held at R-Ranch and Keno Eddy. In June, there continued to be decreased mortality in the groups held at R-Ranch and Keno Eddy compared with other exposure sites, and mortality increased at Tully Creek compared with the May exposure. In September, rainbow trout were exposed at all locations but were held at 18°C, the average river temperature, when returned to the laboratory. This contributes to the overall increased mortality for this exposure. However, mortality at Keno Eddy continued to be low and mortality at R-Ranch was lower than at all sites below Iron Gate Dam.

The cumulative mortality from *C. shasta* for all groups exposed at Beaver Creek in May, June and Sept and held at two temperatures, 13° C and 18° C, is compared in **Figure 3**. For rainbow trout, although mortality was nearly 100% at either temperature each month, the mean day to death was lower for the fish held at 18° C (e.g. 22 d versus 37 d at 13° C in May). For the May and June exposures, mortality in coho salmon was approximately 80% when fish were held at average ambient river temperatures and decreased to

approximately 30% in September. Mortality in Chinook salmon increased from 27% in May to 40% in June, then decreased to less than 5% in September.

Chinook salmon were held at two temperatures at all sites, with mortality predictably higher in the cohorts held at ambient river temperature. There was no mortality in Chinook salmon held in the upper Klamath River, despite the high infection prevalence among rainbow trout held at the Williamson River site. Mortality in Chinook salmon groups held at 13°C was less than 5% at all exposure sites. Comparison of cumulative mortality for groups held at 18°C for each month is shown in **Figure 4**. Mortality occurred in Chinook salmon held at Seiad Valley and Beaver Creek during each exposure period, with the highest mortality occurring in June: Seiad Valley - 40%; Beaver Creek - 30%. At Orleans, mortality was below 5% in May and June and no fish were lost in September. There was no Chinook salmon mortality at any other sites.

Coho salmon were only held at the Beaver Creek site (**Figure 3**). Mortality in the groups held at 13°C after exposure was low for all exposure periods. However, mortality in groups held at ambient river temperature $(18 - 20^{\circ}C)$ was greater than 80% in May and June, and 35% in September. At the higher temperature, mortality in coho salmon was greater and mean day to death lower than for Chinook salmon in all exposures.

Trinity River steelhead exposed at Beaver Creek during June and held at either temperature did not become infected.

Figure 1. Map of sites for exposures of sentinel fish and collection of water samples in 2007.





Figure 2. Mortality curves for susceptible rainbow trout held at all Klamath River sites.

June 2007 Rainbow Trout-13C







Figure 3. Percent mortality for all species exposed at Beaver Creek in May, June and September 2007





Figure 4. Cumulative mortality for Chinook salmon held at exposures sites in the Klamath River mainstem below Iron Gate Dam. Site codes are: KBC – above Beaver Creek, KSV – Seiad Valley and KOR – Orleans



May 2007 Chinook Cumulative Mortality - 18C







September 2007 Chinook Cumulative Mortality - 18C

WATER SAMPLING:

Water samples were taken to determine:

- 1. How infection levels this year compare with levels in previous years.
- 2. If the distribution of the parasite has changed.
- 3. How point sampling compares with longer duration sampling.
- 4. The relationship between parasite numbers measured in water samples and biological effects in the different fish species.

Three types of water samples were taken in 2007 (see Figure 1 for locations):

- 1. Sentinel water was sampled from beside the fish cages at the 7 mainstem sentinel exposure sites: Williamson River, Keno eddy, R-Ranch, Beaver Creek, Seiad Valley, Orleans and Tully Creek. 4 x 1L samples per site were taken when the fish were placed in the river (='in') and when they were removed (='out') in May (15 & 18), June (19 & 22), and September (11 & 14).
- ISCO water was sampled from beside the fish cages at 2 sentinel sites, Beaver Creek and Orleans, for 2-3 consecutive 24 hour periods (500mL every 2 hrs for 24 hours) during the fish exposure period. Both sites were sampled on 16-17 May; 20-22 June; Beaver Creek only 12-14 September.
- 3. Tribal these samplings (through collaboration with the Karuk and Yurok tribes) occurred at 5 mainstem (R-Ranch, Beaver Creek, Seiad Valley, Orleans and Tully Creek) and 4 tributary (Salmon, Scott, Shasta and Trinity Rivers) sites every two weeks from May 10 through September 26 and overlapped in general location with the sentinel sites.

Three of the 4 1L samples collected at each site were tested for inhibition/*Parvicapsula minibicornis* (combined multiplex QPCR) followed by *Ceratomyxa shasta* (singleplex QPCR). Each data point on a graph represents the average of the 3 water samplings at that time point.

Levels of *C. shasta* were already around 10 spores/L at some mainstem sites when sampling commenced in May, 2007 (**Figure 5**). Basin-wide, levels generally peaked at the end of May/beginning of June. Only low levels were recorded in the tributaries at any time of year.

Of the sentinel fish sites (**Figure 6**), Beaver Creek had the highest abundance of spores in May, averaging 10 spores/L or more. This corresponded with the highest cumulative mortality in Chinook salmon. However, the Williamson River had the most spores in June and September, with at least 10 spores/L. This is not reflected in the Chinook exposures. One possible explanation is that species is non-susceptible to the upper river strain of the parasite and this is being investigated. However, rainbow trout exposed at sites with the greatest numbers of parasite present, including the Williamson River, incurred the highest and fastest mortalities. In June, the highest quantities of spores in the lower Klamath River were observed at Beaver Creek and Seiad Valley, and this is again reflected in the highest mortalities in Chinook. Parasite levels increased at all sites from May to June, then decreased by September. The threshold for mortality in Chinook salmon appears to be around the 10 spore/L mark; when less than this number are

detected, cumulative mortality is 10% or less, whereas when this value is exceeded, cumulative mortality is 30% and above.

Levels of *P. minibicornis* closely paralleled those for *C. shasta*, with numbers per liter highest near Beaver Creek. Interestingly, both parasites were detected from tributaries at low levels; however, *P. minibicornis* was less prevalent than *C. shasta* (Figure 5).

Figure 5. Abundance of myxozoan parasites in 11 water samples collected from May through September 2007 by the tribes at 5 mainstem and 4 tributary sites. Each data point is the average of 3 1L samples.



Figure 6. Abundance of *Ceratomyxa shasta* spores in 1L water samples collected in May, June and September by all three methods. Each data point is the average of 3 1L samples.



Collection site



Abundance of Ceratomyxa shasta in the Klamath

Collection site