California Nevada Fish Health Center
Joint USFWS USGS Technical Report:

Pilot study to access the role of *Ceratomyxa shasta* infection in mortality of fall-run Chinook smolts migrating through the lower Klamath River in 2008.

USFWS: Scott Foott*, Greg Stutzer**, and R. Fogerty.
USGS: Hal Hansel, Steven Juhnke, and John Beeman ++

*   direct correspondence
**   USFWS, Arcata Fish & Wildlife Office, Arcata, CA
++   U.S. Geological Survey, Western Fisheries Research Center, Columbia River Research Laboratory, Cook, WA

Summary: Apparent survival and migration rate of radio-tagged hatchery subyearling Chinook salmon released at Iron Gate Hatchery was monitored in the Klamath River to see if the timing of mortality coincided with observations of ceratomyxosis in re-captured coded wire tag cohorts. Despite rapid emigration, these relatively large (mean fork length 92 mm) smolts had a cumulative apparent survival to the estuary of 0.074 (SE 0.024) and standardized rates of survival per 100 km tended to decrease linearly with distance from the hatchery. The last fish detection occurred 26 days after release but median travel time between Iron Gate Hatchery (rkm 309) and the last receiver near the Klamath estuary (Blake's Riffle rkm 13) was about 10 days. The majority of apparent mortality (8-10 d post-release) occurred before disease from *Ceratomyxa shasta* infection is expected after exposure to infectious waters. Despite numerous observations of ceratomyxosis in the Klamath R. during June, an obvious link between disease and apparent survival was not present in this study. Future studies should examine the acute (e.g., predator types and densities) and chronic (e.g., swimming performance impairment due to disease) mortality factors for juvenile Chinook salmon smolts in the Klamath River.

Introduction: Juvenile salmon in the Klamath River incur a high incidence of infection from two myxozoan parasites, *Ceratomyxa shasta* and *Parvicapsula minibicornis* (Stocking et al. 2006, Foott et al. 2004). A section of the lower Klamath River has been identified to be highly infectious (Stocking et al. 2006, Hallett and Bartholomew 2006). The incidence of *C. shasta* infection, observed in histological sections of juvenile Chinook salmon collected in the Klamath River above the confluence of the Trinity River between May and July, has ranged from 21–35% (Nichols et al. 2008). Over 90% of these fish are also infected with the kidney parasite *P. minibicornis.* Approximately 70% of the positive histology samples demonstrated pathology due to the infection. Similarly, sentinel salmon studies have demonstrated $\geq$ 80% mortality due to parasitic disease after 3-day exposures in the Klamath River (Stone et al. 2008). The high prevalence and severity of myxozoan infection in native fish that should have high resistance to this endemic disease indicates these parasites may be limiting salmon recovery in the Klamath River. These parasites occur in a number of Pacific Northwest watersheds and the life cycles of both parasites include the polychaete, *Manayunkia speciosa,* as an alternate host (Bartholomew et al. 1997, Bartholomew et al. 2006). The actinospore, a stage that is infectious to salmon, is released from infected polychaetes into the water column. Infections by *C.shasta* can occur from spring through fall at water temperatures $\geq$ 7°C (Ching and Munday 1984, Hendrickson et al. 1989). An unanswered question is how this disease situation affects overall survival of juvenile salmon out-migrating in the Klamath River through both direct and indirect (e.g., increased susceptibility to predation) effects.

The California Department of Fish and Game's Iron Gate Hatchery (IGH) is located at the limit of anadromous fish range (rkm 309) and releases 4-6 million fall-run Chinook (*Oncorhynchus tshawytscha)* smolts during May and June. These fish are the progeny of brood stock collected at the base of Iron Gate Dam. A portion ($\leq$ 6%) of the release group is marked with coded wire tags (CWT) that allow for the determination of "days at large" if recaptured in the river (Nichols et al. 2008). The infection status of these CWT smolts can be used to estimate similar disease impacts on natural smolts rearing and migrating from the

3

river above the confluence of the Scott River such as the Shasta River. Tracking radio-tagged cohorts of IGH release groups should provide data on their migration rate and reach-specific survival. A unique opportunity occurred in 2008 in which funding for radio tags coincided with the deployment of radio-tag monitoring infrastructure and expertise in the lower Klamath River for a similar USGS-USFWS project on juvenile coho salmon. Funding for the study came from a settlement agreement over effluent impacts from Iron Gate Hatchery and was supplied by a co-defendant (PacifiCorp).

The objectives of this study were to 1) determine reach-specific survival and migration timing of radio-tagged fall-run Chinook juveniles released with IGH production groups in the spring, and 2) compare infection data from recovered coded wire tagged IGH smolts and other concurrent fish health studies with radio-tag cohort survival. The goal of this study was to determine if the time frame of mortality in fish migrating in the Klamath River with known migration histories was similar to that of fish from concurrent fish health studies, and thus provide a measure of the effect of disease in mortality of river migrants.

Methods:
Study site – The study site included the Klamath River from Iron Gate Hatchery (river km 309) to Blake's Riffle (river km 13).   The timing of passage was monitored at eight downstream sites until August 27, 2008 (Figure 1). The duration of the study was determined by an independent investigation at the USGS Columbia River Research Laboratory that determined the maximum tag life expectancy to be 77 days.

Fish – The production fish at the hatchery are too small for implanting radio tags, so a subset of them was placed on an accelerated growth regime for this study. Two hundred and fifty juvenile IGH Chinook salmon (mean wt. 1.46 g) were taken to the California Nevada Fish Health Center (FHC) wet lab on April 4, 2008. Water temperature at IGH was 9° C on the day of transport. The fish were reared in an 800 L circular tank supplied with 5.7 L / min flow of 10°C water and aeration. The lab receives ozonated Coleman National Fish Hatchery water and
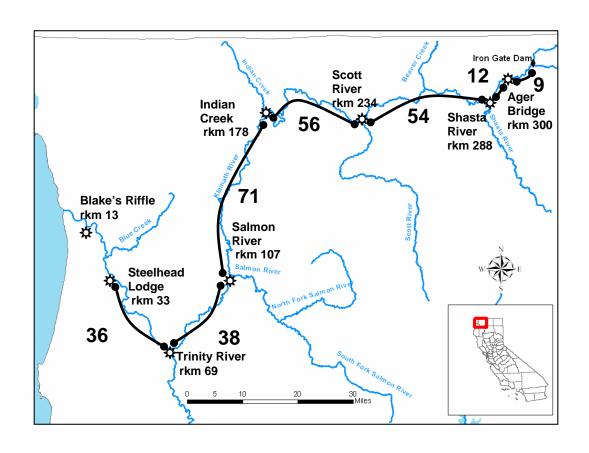
4

Figure 1. Study area of the lower Klamath River subyearling Chinook salmon survival study, northern California, 2008. Fish were released at Iron Gate Fish Hatchery (rkm 309) and monitored at downriver detection sites indicated by a ☼. Bold numerals indicate reach lengths in kilometers. Figure modified from U.S. Fish and Wildlife Service, Arcata, California, 2006.

there is no history of *C. shasta* or *P. minibicornis* infection at this facility. Water temperature was increased to 16°C over an 18 d period and Bio-Oregon salmon diet (#1) was fed at 8% body weight / d. Sizes of 20 salmon were measured every week. Daily growth rate ranged from 0.61 to 1.17 mm/d over the 60 d rearing period. The largest 204 salmon were taken back to IGH on June 6 and 175 fish of proper size were tagged on June 10.

Tag implantation – Radio tags were surgically implanted at IGH. The tags weighed 0.43 g in air and were 13 mm long, 5 mm wide, and 3 mm high with a 16 cm long trailing antenna. After the surgery, fish were allowed to recover 24 hours prior to release. Radio transmitters (NTC-M-2; Lotek Wireless Inc,

Newmarket Ontario Canada) were identical to functional transmitters used in previous survival studies (Beeman et al. 2007, 2009). Procedures for surgical implantation of radio transmitters were similar to those described by Adams et al. (1998). Prior to insertion, transmitters were disinfected using a 0.5% solution of chlorohexidine diacetate (Nolvasan® Fort Dodge Animal Health, Fort Dodge, Iowa). Transmitters were rinsed twice in sterile water and placed on the sterile portion of a surgical glove wrapper along with the surgical instruments immediately before surgery. Two sets of surgical instruments were used enabling one set to be disinfected by soaking in 70% ethanol while the other set of instruments was being used in surgery. To implant the transmitter, a 7-mm (approximate) incision deep enough to penetrate the peritoneum was made about 5 mm anterior to the pelvic girdle and about 3 mm away from and parallel to the mid-ventral line (Summerfelt and Smith 1990). The shielded-needle technique described by Ross and Kleiner (1982) was used to provide an outlet through the body wall for the transmitter antenna. The transmitter was positioned to lie slightly posterior to the incision by gently pulling on the antenna. A single simple interrupted suture (Ethicon coated vicryl, 5-0 reverse cutting P-3 needle) closed the incision. The target weight for tagging was 8.6 g or greater to result in a tag-weight to body-weight ratio of 5% (Adams et al. 1998, Hall et al. 2009). Three-to-four tagged salmon were held in 5 gal ventilated buckets with lids that were placed into a hatchery raceway. Twenty tags were used for tag life analysis. The tagged fish were released into the Klamath River directly adjacent to Iron Gate Hatchery at 2140 hours on June 11. Only 121 fish of the 175 (70%) tagged fish were successfully released due to a problem with tangling of the antennas within the buckets. Fifteen of the 121 released fish required extra handling to untangle their antenna.

   Data analysis - Both unstandardized and standardized apparent survivals of radio-tagged subyearling Chinook salmon were estimated for the reach above each of the eight detection sites (Figure 1). Reach names used in this report refer to the detection site at the downstream end of each reach. Reach survivals were estimated with Cormack-Jolly-Seber (CJS) capture-mark-recapture methods (Cormack, 1964; Jolly, 1965; Seber, 1965). Apparent survival is the

probability that an animal is available for recapture and is a function of losses to mortality, emigration outside the spatial and temporal bounds of the study, or permanent cessation of movement within the study boundaries. Since the processes of emigration outside the study area and cessation of migratory behavior could not be quantified, they are treated here as losses due to mortality. Unstandardized survival rates in this report are synonymous with apparent reach survivals unadjusted for varying reach lengths. Standardized survival rates refer to apparent reach survivals scaled to a common reach length (100 km), making relative comparisons among reaches more straightforward.   Cumulative apparent survival from IGH to Blake's Riffle was estimated as the joint probability of the individual unstandardized reach survivals with the standard error calculated using the delta method (Seber 1982).

Results and Discussion:

Release - Over 5.3 million juvenile Chinook salmon (brood year 2007) were released from IGH between May 27 and June 16, 2008 (4.98 million unmarked and 305,700 CWT fish). Average fish weight for each release group ranged from 4.6 to 5.6 g. The radio-tagged group was released with an 876,000 production lot on June 11. This was the third of four releases for the hatchery. Average weight (9.27 g, SD 1.25 g) and fork length (92 mm, SD 4 mm) of the radio-tagged group was substantially larger than the production cohorts (40-50% heavier), and as such they did not strictly represent the untagged fish. The antennas of some fish (54 of 175) tangled during post-tag holding and those fish could not be used in the study. The tag weight-to-body weight ratio of the 121 fish released ranged from 3.2 to 5.5%, with an average of 4.8%. The median fork length of juvenile Chinook salmon captured for parasite assays during June 2008 in the Klamath River between the Shasta and Trinity rivers was 76 mm (Nichols et al. 2009). The median fork length of the radio-tagged fish released was 91 mm.
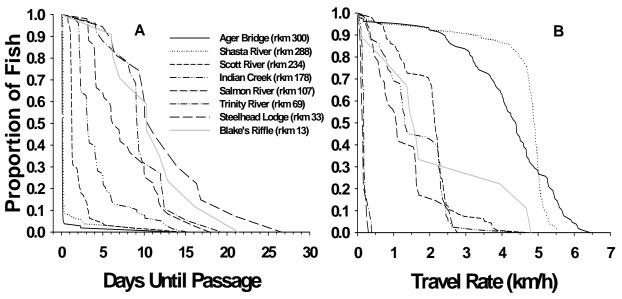
Figure 2. Travel time from release (A) and travel rate (B) for eight lower Klamath River reaches. Travel times were calculated from time of release to last detection at the downstream detection site of each reach. km/h = kilometers per hour.

Migration timing - The last fish detection at any of the eight reaches occurred on July 8, 26 days after release. Between the release date and the last detection, water temperature increased from about 15 to 22° C and river flows decreased from 1,990 to 1,020 cubic feet per second below Iron Gate Dam and from 12,800 to 6,280 cubic feet per second at Blake's Riffle. The median travel time to rkm 13 was 10.2 days (N = 9, Figure 2A). Wallace (2004) reported that median travel times (as measured by median DAL values) for coded wire tagged juvenile IGH Chinook released between 1993 and 2001 ranged from 26-52 days. In 2001, median DAL for IGH smolts captured in the estuary (average fork length 92.5 mm) was 32 d. This size was similar to the study group (92mm), but travel time was approximately 3 times longer. Median travel rates through the upper two reaches (Ager Bridge and Shasta River) were greater than 4.4 km/h, at least twice as fast as the median travel rates for successive downriver reaches which generally decreased with increasing distance from the hatchery (Figure 2B). Travel rates were lowest for the Trinity River and Steelhead Lodge reaches (< 0.5 km/h). These differences in travel rates were in part explained by the release time, or the time that a reach was entered and its length. Ninety percent of the detections past all the detection sites, independent of the reach, occurred

between 2100 hours and 0600 hours. Following release at 2100 hours, most fish moved quickly through the first two reaches, passing the Shasta River detection array before sunrise the next day. Fish were generally within the 54 km Scott River reach when they encountered the first daylight period (about 16 h) and stopped migrating until evening, or at least did not pass detection sites until evening. If fish migrated primarily at night, those traversing this reach and reaches downstream would have reduced travel rates relative to the Ager Bridge and Shasta River reaches because they encountered more hours of daylight. Increasing water temperature could have also delayed migration rate if fish were holding in thermal refugia present in some reaches and not in others.

Survival estimates of radio-tagged fish- Four single-release CJS models were developed and evaluated to estimate survival and recapture (detection) probabilities from IGH to Blake's Riffle (Table 1). Each model included a separate set of parameters for survival and recapture probabilities that corresponded to one of two hypotheses. The first hypothesis assumed no difference in survival or recapture probabilities among reaches, whereas the second hypothesis assumed that there were differences among reaches. All possible combinations of the two hypotheses for the survival and recapture probability parameters were represented in the four models evaluated. The degree to which the data supported each model was determined using Program MARK (White and Burnham, 1999). This program derives individual model parameter estimates and confidence intervals from the mark-recapture data and calculates the Akaike Information Criterion for small sample sizes (AICc) and other statistics used to rank the competing models. The model or models best supported by the data were determined by examining the AICc values, number of parameters, and deviances relative to the other proposed models as described by Burnham and Anderson (2002).

The most parsimonious model for the subyearling Chinook salmon mark-recapture data assumed differences in survival among reaches, but no differences in recapture probabilities (Table 1). This model received 99.8% of the AICc weights and was 500 times more likely than the next best model in the

set, indicating that there was little uncertainty as to which combination of hypotheses were most supported by the data. The competing models received virtually no support and the corresponding hypotheses were highly improbable.

Table 1. Model summary from analyses of unstandardized apparent survival (Phi) and recapture probabilities (P) to estimate reach survivals of subyearling Chinook salmon in the lower Klamath River, northern California, 2008.

[Models are based on data from 121 hatchery fish released on 11 June 2008 at Iron Gate Fish Hatchery. Model descriptions include factors allowed to vary within survival (Phi) and recapture (P) probabilities. Models are ranked based on AICc, a modification of Akaike Information Criterion for small samples. A '.' indicates that no reach effects were assumed (i.e. a single estimate was derived for all observations).]

Model rank	Model	AICc	Delta AICc	AICc weight	Model likelihood	Number of parameters	Deviance
1	Phi(reach), p(.)	475.571	0.000	0.998	1.000	9	457.254
2	Phi(reach), p(reach)	488.107	12.536	0.002	0.002	15	457.254
3	Phi(.), p(.)	572.794	97.223	0.000	0.000	2	568.773
4	Phi(.), p(reach)	584.981	109.411	0.000	0.000	9	566.665

The estimates of survival from the top-ranked model reflect the differences in unstandardized apparent survival among reaches that are supported within the data. The individual reach survival estimates ranged from 0.488 (standard error [SE] 0.078) for the Trinity River reach to 1.000 (SE 0.000) for the Shasta River reach (Table 2). The estimated common recapture probability for all reaches was 1.000 (SE 0.000). This common recapture probability uniquely allows survival to be estimated for Blake's Riffle even though there were no detection sites further downstream from which to directly estimate the recapture probability for this site. In cases where modeling indicates heterogeneity among recapture probabilities, the survival and recapture probability for the last reach are not separable. Based on these reach survival estimates, cumulative survival generally decreased with distance from IGH (Figure 3). The cumulative apparent survival and 95% confidence interval for the entire reach from IGH to Blake's Riffle was 0.074 ± 0.047 (Table 2).

Table 2. Estimated unstandardized apparent survivals and profile likelihood confidence intervals for subyearling Chinook salmon in eight reaches of the lower Klamath River, northern California, 2008.

[Results are based on the most parsimonious model (Table 1) supported by mark-recapture data from 121 radio-tagged hatchery fish released near Iron Gate Fish Hatchery on 11 June 2008 at 2100 hours. Survival estimates are not standardized for reach length. Confidence intervals for cumulative estimates are not profile likelihood.]

Reach number	Reach description	Reach length (km)	Survival estimate	Standard error	95% confidence interval	
					Lower	Upper
1	Hatchery to Ager (rkm 309 to 300)	9	0.868	0.031	0.800	0.920
2	Ager to Shasta River (rkm 300 to 288)	12	1.000	0.000	0.982	1.000
3	Shasta River to Scott River (rkm 288 to 234)	54	0.905	0.029	0.839	0.951
4	Scott River to Indian Creek (rkm 234 to 178)	56	0.821	0.039	0.736	0.889
5	Indian Creek to Salmon River (rkm 178 to 107)	71	0.526	0.057	0.415	0.634
6	Salmon River to Trinity River (rkm 107 to 69)	38	0.488	0.078	0.339	0.638
7	Trinity River to Steelhead Lodge (rkm 69 to 33)	36	0.700	0.102	0.483	0.868
8	Steelhead Lodge to Blake's Riffle (rkm 33 to 13)	20	0.643	0.128	0.383	0.854
	Hatchery to Steelhead Lodge	276	0.114	0.029	0.058	0.171
	Hatchery to Blake's Riffle	296	0.074	0.024	0.027	0.120

Standardized subyearling Chinook salmon survival rates generally decreased linearly downriver from IGH to Blake's Riffle (Figure 4). The survival rate from Ager Bridge to Shasta River was 1.0 per 100 km, whereas the survival rate in the last reach, from Steelhead Lodge to Blake's Riffle, was 0.11 per 100 km. The one exception to this was the 9 km reach between the release site and Ager Bridge (0.21 per 100 km), which was more similar to the reaches furthest downriver than the other reaches (Figure 4).

The acute rate of mortality in the first reach may be indicative of an initial set of fish behaviors or conditions that are unique to the tagged fish immediately after release. For example, tagged fish may be more disoriented or naive immediately after release making them more vulnerable to predation, or otherwise more ill-suited to the rigors of in-river survival after being reared in the hatchery, surgically implanted with a tag, handled, and released. Alternatively, the relatively low survival may not reflect initial fish behavior after release, but instead, differences in predator densities near the hatchery and dam, or some other reach-specific characteristic that results in an increased mortality rate. The release occurred after three previous large production releases which could have
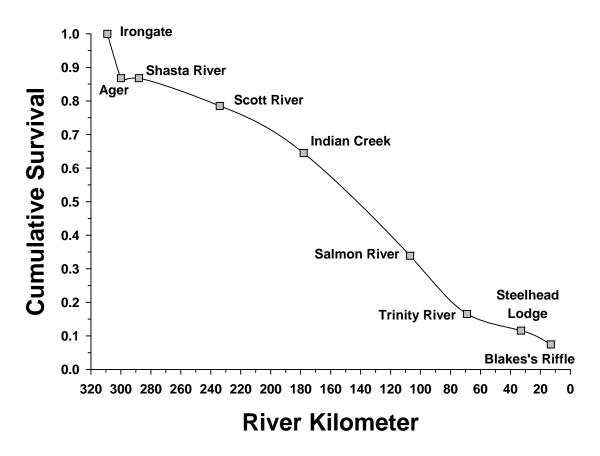
Figure 3. Cumulative apparent survival of radio-tagged subyearling Chinook salmon from release at Iron Gate Fish Hatchery to eight downriver detection sites on the lower Klamath River, northern California, 2008. Cumulative survivals were calculated as the product of the individual reach survival estimates in Table 2.

increased predator density in this reach. Shively et al. (1996) reported that northern pikeminnow (*Ptychoceilus oregonensis*) rapidly shifted to a juvenile salmon diet following a hatchery release, indicating predators can respond to hatchery releases. Either, or both, of these alternatives could explain the acute mortality rate. Mortality predominantly due to fish behavior associated only with tagging and handling would cause the apparent survival to be an inaccurate representation of the true survival.

Except for release to Ager Bridge as noted above, the otherwise linear decrease in survival rates among reaches (Figure 4) with distance could be the result of chronic or acute effects on survival. Examples of potential chronic factors include long-term tag effects, disease, or exposure to environmental
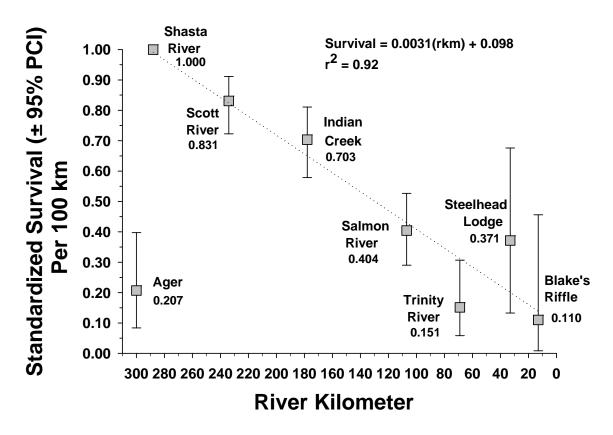
Figure 4. Lower Klamath River subyearling Chinook salmon apparent survival rates adjusted for reach length versus river kilometer, 2008. Survival rates have been standardized for a 100 km reach length. Regression does not include data from Ager.

factors such as high water temperature. The effects of fish holding between detection sites or entering tributaries could also depress the survival as estimated in this study, but we did not measure these factors. Examples of potential acute mortality effects include predator densities or some other environmental factor that increased or decreased linearly downriver and contributed to the mortality rate. Few fish detected at Indian Creek were subsequently detected at the Salmon River, prompting the USFWS to snorkel this reach between July 9 and July 11; twelve tags were recovered. The source of this mortality was not determined.

There did not appear to be a link between mortality of radio-tagged fish and disease (ceratomyxosis), based on travel times of radio-tagged fish and known progression rates of ceratomyxosis. The timing of most estimated

13

mortality of radio-tagged fish occurred prior to when we would expect impairment due to ceratomyxosis. Of the 21 fish detected at the Trinity River receiver, 15 (71%) were detected ≤ 10 d post-release. In previous sentinel salmon studies in the Klamath River, ceratomyxosis morbidity and associated tissue lesions were observed beginning 10 – 14 days post-exposure (Foott et al. 2004, Stone et al. 2008). A similar onset of clinical ceratomyxosis occurred in a June 2008 sentinel study described below (CA-NV FHC Prognosis study data, report pending). We hypothesize that disease morbidity would increase mortality (reduced apparent survival) in this 10 -14 d post-release period. The linear decrease in survival rates with distance and time did not demonstrate this effect; however, disease could have affected mortality indirectly. For example, Mesa et al. (1998) reported that Chinook salmon with clinical levels of Bacterial Kidney Disease (as measured by relatively high levels of *Renibacterium salmoninarum* antigen) experienced higher predation than control fish. It is also possible that disease progressed faster in the tagged fish than in untagged fish used in previous sentinel studies.

Observations of ceratomyxosis in untagged juvenile salmon - In the period between May and July 2008, the incidence of *C. shasta* infection in juvenile Chinook salmon collected in the Klamath River above the confluence with the Trinity River was 38% by histology and 49% by the more sensitive QPCR assay (Nichols et al. 2009).   Signs of disease were seen in > 60% of the intestine sections containing the *C. shasta* parasite. In early June 2008, juvenile coho and Chinook salmon sentinels were exposed in the Klamath River (approximately rkm 265) for 72 h and transported back to the FHC wetlab for extended rearing at 18°C. These fish experienced over 90% cumulative mortality due to ceratomyxosis within 24 days post-exposure (CA-NV FHC Prognosis study data, report pending). These data indicate that the lower Klamath River was highly infectious for *C. shasta* in June 2008.

Incidence of C. shasta infection in CWT smolts - A total of 119 CWT Chinook salmon smolts from IGH were collected in the 2008 Klamath River salmon monitoring project with the majority captured below the confluence of

Trinity River (Nichols et al. 2009). Of the 32 smolts diagnosed by QPCR to be *C. shasta* positive (27%), four of these fish were from the June 11 release (same as radio tag cohorts). All four June 11 recaptures occurred near Metah Creek (rkm 57.7, between Steelhead Lodge and Trinity River detection sites) between 16 and 21 days post release.

       In-river tracking of radio-tagged fish - A float survey was conducted from Indian Creek (rkm 178) and Green Riffle (~rkm 120) between July 9-11 to locate 39 tags that passed the Indian Creek receiver but were not detected at the Salmon River (rkm 107) receiver. The 12 radio tags within this reach that were detected and recovered were evenly dispersed along the river continuum and were most often located in low gradient habitat types along eddy shear zones in water ranging from <1 to 3 m in depth. While diving to locate these transmitters, a number of dead juvenile salmonids and other fish species were observed. Although no formal counts were conducted, the crew observed about 10 to 40 dead fish within 5 meters upstream and downstream of the location of each tag. Dead fish were observed during 23 of the 25 dives. The appearance of dead fish observations ranged from what was assumed to be recent mortalities to carcasses fully covered with fungus. The two dives where dead fish were not observed occurred in water having relatively high water velocity. Dead and / or dying fish were also observed at several thermal refugia areas, which were also occupied by live salmonids (predominantly Chinook salmon, and to a lesser extent, steelhead *O. mykiss*). Most mortalities observed were juvenile salmonids; however, numerous dead sculpins (*Cottus* sp.), suckers (*Catostomus* sp.), and one dead bullhead (*Ictaluras* sp.*)* catfish were also observed.

Conclusions:

       We determined the apparent survival of radio-tagged hatchery subyearling Chinook salmon from a single bulk release on June 11, 2008 and monitored downriver detection sites for these fish through August 27. Despite rapid emigration of these relatively large subyearling fall Chinook salmon, cumulative apparent survival for the entire 296 km river segment was low (0.074 SE 0.024) and standardized rates of survival per 100 km tended to decrease linearly with

distance from the hatchery. The cumulative apparent survival of radio-tagged fall Chinook salmon over the 276 km from release to Steelhead Lodge (rkm 33) in this study was 0.114 (SE 0.029). Radio-tagged juvenile coho salmon migrating seaward through this 276 km area earlier in the spring of 2008 (chiefly in April and May) had cumulative apparent survivals of 0.290 (SE 0.056) for hatchery fish captured in an in-river trap and 0.406 (SE 0.032) for those taken directly from Iron Gate Hatchery (Beeman et al. 2009).

Several limitations in this study may have affected the results. The methods used could not separate the various sources of true mortality, such as predation and disease, from those that were assumed to be mortality, such as movement into tributaries and cessation of migration between detection sites. In addition, the tagged fish in this study were grown to a size much larger than the untagged fish released concurrently at the hatchery so they would be suitable for the radio transmitter. As such, the tagged fish in this pilot study may not have represented the migration or survival of the untagged fish released. These factors make it likely that the apparent survival estimated in this study does not represent the true survival of untagged fish. The true survival of untagged juvenile fall Chinook salmon in the lower Klamath River remains unknown. However, representing untagged fish was not the primary goal of this study: the goal was to determine if mortality of fish migrating in the river occurred during a similar timeframe as mortality in fish from sentinel studies and could therefore be attributed to disease.

In studies of tagged fish such as this one, we can know where mortality occurred, but rarely can we know much about when it occurred. This is because we generally only detect live fish when using automated detection stations. As such, we can state that a fish likely died between specific detection stations and sometime after its last detection, but we know little else about the when death occurred. We only know the minimum life of each tagged fish. One could assume that the migration timing of fish that lived and those that died were similar, and from that infer more about when fish died. We did not test this assumption, and if false, we can infer little about the timing of death and its

relation to disease other than it likely occurred sometime after their last detection. If there had been few mortalities until 10-14 days after release we may have been able to infer a role of disease, but this did not happen. Assigning the time of death more accurately may be possible if tagged fish were used with the addition of frequent mobile tracking trips.

Future studies on smolt survival should identify the types and densities of predators in the Klamath as well as the use of thermal refugia and tributaries by smolts. The Klamath River was highly infectious for *C. shasta* in June 2008, however, no strong association with disease and apparent survival could be made with the radio-tagged group. A large percentage of apparent mortality occurred prior to when clinical ceratomyxosis would be expected in the smolts. The effect of early inflammatory changes associated with initial infection by *C. shasta* and *P. minibicornis* on fish performance is not known and could influence susceptibility to predation. Conversely, rapid emigration through the infectious zone in the Klamath River (Stocking et al. 2006) could reduce the potential for disease and separate the study group from other ceratomyxosis observations in juvenile Chinook salmon during June.

The last fish detected passing any of the eight detection sites occurred on July 8, 26 days after release, but the median travel time between the hatchery and Blake's Riffle was about 10 days. Fish size in the tag group was larger than both the hatchery release and juvenile Chinook salmon captured in health monitoring efforts in June. We hypothesize that emigration rate would be longer for these smaller fish and could expose them to higher acute (predation) and chronic (disease) mortality factors than the study group. This initial effort in radio tag monitoring provided data for a brief snap shot in time during the outmigration period that can extend over several months (Wallace 2004). Future work should examine the infection and disease response of juvenile salmon held in traditional sentinel cages (fixed locations within the infectious zone) to cohorts that are moved through this zone at a rate comparable to that observed in the current study. It would also be useful to study the effects of *C. shasta* and *P. minibicornis* on susceptibility to predation.

Contribution of authors:

USFWS:

J. Scott Foott - study proposal, tagging assistance, editing of joint report

Greg Stutzer - tagging and release of study group, in-river tag recovery

 Ryan Fogerty - rearing of study group, tagging assistance

USGS:

 Hal Hansel - survival and migration data analyses and summary

 Steven Juhnke - tagging assistance and release of study group, data management

John Beeman - study proposal, editing of joint report

References:

Adams, NS, DW Rondorf, SD Evans, JE Kelly, and RW Perry. 1998. Effects of Surgical and gastrically implanted radio transmitters on swimming performance and predator avoidance of juvenile Chinook salmon (*Oncorhynchus tshawytscha*). Canadian Journal of Fisheries and Aquatic Sciences 55:781-787.

Bartholomew JL, SD Atkinson, and SL Hallett. 2006. Involvement of *Manayunkia speciosa* (Annelida: Polychaeta: Sabellidae) in the life cycle of *Parvicapsula minibicornis,* a myxozoan parasite of Pacific salmon. Journal of Parasitology 92:742-748.

Bartholomew JL, MJ Whipple, DG Stevens, and JL Fryer. 1997. The life cycle of *Ceratomyxa shasta*, a myxosporean parasite of salmonids, requires a freshwater polychaete as an alternative host. Journal of Parasitology 83(5):859-868.

Beeman, JW, SD Juhnke, GM Stutzer, and NJ Hetrick. 2009. Survival and migration behavior of juvenile coho salmon in the Klamath River relative to discharge at Iron Gate Dam, 2008. U. S. Geological Survey Open-file report 2009-1019. Available online: http://pubs.usgs.gov/of/2009/1019.

Beeman, JW, GM. Stutzer, SD Juhnke, NJ Hetrick. 2007. Survival and migration behavior of juvenile coho salmon in the Klamath River relative to discharge at Iron Gate Dam, 2006. Final report prepared by U. S. Geological Survey, Cook, Washington and U. S. Fish and Wildlife Service, Arcata, California for the U. S. Bureau of Reclamation, Mid-Pacific Region, Klamath Basin Area Office, 06AA204092 and 07AA200181, Klamath Falls, Oregon.

Burnham, K.P., and D.R. Anderson. 2002. Model selection and multimodel inference: A practical information-theoretic approach: Springer Verlag, 488 p.

Ching, HL, and DR Munday. 1984. Geographic and seasonal distribution of the infectious stage of *Ceratomyxa shasta* Noble, 1950, a myxozoan salmonid pathogen in the Fraser River system. Canadian Journal of Zoology 62:1075-1080.

Cormack, RM. 1964. Estimates of survival from the sighting of marked animals. Biometrika 51:429-438.

Foott JS, R Harmon, and R Stone. 2004. Effect of water temperature on non-specific immune function and Ceratomyxosis in juvenile Chinook salmon and steelhead from the Klamath River. California Fish and Game 90(2):71-84.

Hall. JE, J Chaberlin, AN Kagley, C Greene and KL Fresh. 2009. Effects of gastric and surgical insertions of dummy ultrasonic transmitters on juvenile Chinook salmon in seawater. Transactions of the American Fisheries Society 138:52-57.

Hallett SL and JL Bartholomew. 2006. Application of real-time PCR assay to detect and quantify the myxozoan parasite *Ceratomyxa shasta* in water samples. Diseases of Aquatic Organisms 71:109-118.

Henderickson GL, A Carleton, and D Manzer. 1989. Geographic and seasonal distribution of the infective stage of *Ceratomyxa shasta (*Myxozoa) in Northern California. Diseases of Aquatic Organisms 7:165-169.

Jolly, GM. 1965. Explicit estimates from capture-recapture data with both death and immigration-stochastic model. Biometrika 52:249-259.

Mesa MG, TP Poe, AG Maule, and CB Schreck. 1998. Vulnerability to predation and physiological stress responses in juvenile Chinook salmon (*Oncorhynchus tshawytscha*) experimentally infected with *Renibacterium salmoninarum*. Canadian Journal of Fisheries and Aquatic Sciences 55:1599-1606.

Nichols K, K True, R Fogerty, L Ratcliff and A Bolick. 2009. Myxosporean Parasite (*Ceratomyxa shasta* and *Parvicapsula minibicornis*) prevalence and severity in Klamath River Basin juvenile Chinook and coho salmon, April-August 2008. U.S. Fish and Wildlife Service, California-Nevada Fish Health Center, Anderson, CA. Available online: http://www.fws.gov/canvfhc/reports.asp

Nichols K, K True, R Fogerty and L Ratcliff. 2008. FY 2007 Investigational Report: Klamath River juvenile salmonid health monitoring, April-August 2007. U.S. Fish & Wildlife Service California-Nevada Fish Health Center, Anderson, CA. Available online: http://www.fws.gov/canvfhc/reports.asp.

Ross, MR and CF Kleiner. 1982. Shielded-needle technique for surgically implanting radio-frequency transmitters in fish. The Progressive Fish Culturist 44:41-43.

Seber, GAF. 1965. A note on the multiple-recapture census: Biometrika 52:249-259.

Seber, GAF. 1982. The estimation of animal abundance and related parameters. Macmillan, New York. 652 p.

Shivey RS, TP Poe, and ST Sauter. 1996. Feeding response by northern squawfish to a hatchery release of juvenile salmonids in the Clearwater River, Idaho. Transaction of the American Fisheries Society 125:230-236.

Stocking RW, RA Holt, JS Foott, and JL Bartholomew. 2006. Spatial and temporal occurrence of the salmonid parasite *Ceratomyxa shasta* in the Oregon-California Klamath River Basin. Journal of Aquatic Animal Health 18:194-202.

Stone R, JS Foott, and R Fogerty. 2008. FY 2006 Investigational Report: Comparative susceptibility to infection and disease from *Ceratomyxa shasta* and *Parvicapsula minibicornis* in Klamath River basin juvenile Chinook, coho and steelhead populations. U.S. Fish and Wildlife Service California-Nevada Fish Health Center. Anderson, CA. Available online: http://www.fws.gov/canvfhc/reports.asp

Summerfelt, RC and LS Smith. 1990. Anesthesia, surgery, and related techniques. Pages 218-272 *in* CB Schreck and PB Moyle, editors. Methods for Fish Biology. American Fisheries Society, Bethesda, Maryland.

Wallace, M. 2004. Natural vs. hatchery proportions of juvenile salmonids migrating through the Klamath River Estuary and monitor natural and hatchery juvenile salmonid emigration from the Klamath River Basin: July 1, 1998 through June 30, 2003. California Department of Fish and Game Inland and Anadromous Sport Fish Management and Research, Klamath River Basin Juvenile Salmonid Investigations. Final performance report - Federal Aid in Sport Fish Restoration Act - Project No. F-51-R-6, Arcata, CA. 50 p.

White, GC and KP Burnham. 1999. Program MARK: survival estimation from populations of marked animals: Bird Study 46 Supplement:120-138.