Assessing Thiamine Content of Klamath River Basin Chinook Salmon

By

NATHAN YANCHEFF
CAPSTONE PROJECT

Submitted in adherence to the curriculum requirements for the degree of

MASTER OF ADVANCED STUDIES - MARINE BIODIVERSITY & CONSERVATION

At

SCRIPPS INSTITUTION OF OCEANOGRAPHY, UNIVERSITY OF CALIFORNIA SAN DIEGO

Approved:

Capstone Advisory Committee
June, 16, 2023

Keith Parker, Chair

DocuSigned by:
[redacted]
7B29C8A7B63044B...

Nate Mantua

DocuSigned by:
[redacted]
8798330620434E7...

Thomas Williams

DocuSigned by:
[redacted]
BCE7D6271E1B4B1...

Isabel Rivera-Collazo

DocuSigned by:
[redacted]
5B0545F163184BA...

Abstract

Thiamine Deficiency Complex is an emerging health concern for California's Chinook salmon (*Oncorhynchus tshawytscha*). Low egg thiamine concentrations contribute to the condition being expressed in newly hatched fry leading to increased rates of early mortality. Increased rates of mortality in early life stages could further jeopardize already depleted populations of Chinook salmon in California's river systems. In 2020, a study of egg thiamine concentrations at Coleman National Fish Hatchery and Livingston Stone Fish Hatchery revealed nearly 50% of the sampled populations contained egg thiamine concentrations below 5 nmol/g which is the egg thiamine concentration threshold for 95% fry viability. This study assesses egg thiamine concentrations from Chinook salmon sampled in the Klamath River Basin during their 2020 and 2021 annual spawning migrations. The study will also assess relationships between egg thiamine concentration and other variables including water temperature and distance of migration, and it will also evaluate relationships between egg diameter and egg thiamine concentration.

Introduction

Thiamine is an essential micronutrient which is important for energy and fatty acid metabolism. Fish obtain thiamine from their diet. Thiamine Deficiency Complex (TDC) is a condition triggered by an inability to either acquire or retain sufficient thiamine (vitamin B1). Fish offspring afflicted with TDC exhibit symptoms which include loss of equilibrium, lethargy, swimming erratically, and swimming in spiraling patterns (Riley and Evans 2008; Harder et al. 2018). The condition can result in high mortality due to impaired neurological functioning during the early life stages of fish development (Honeyfield et al. 2005). Fry (i.e., recently hatched juveniles) exhibiting TDC symptoms can be rehabilitated by being immersed in a thiamine bath. Their condition rapidly improves and mortality rates drop when they receive this treatment (Reed et al. 2023). The vitamin is readily available in most ecosystem food chains indicating that a lack of its presence in the diet of afflicted species is likely not the core cause of the condition. Historically, occurrences of TDC have been accompanied by the consumption of prey that contain thiaminase, which is a thiamine degrading enzyme (Harder et al. 2018).

Chinook salmon have historically had a diverse diet including Pacific sardines (*Sardinops sagax*), northern anchovies (*Engraulis mordax*), Pacific mackerel (*Scomber japonicus)*, Pacific herring (*Clupe pallasii)*, market squid (*Doryteuthis opalescens*), krill (*Euphausiacea*), and other low trophic prey species. Anchovies, and many other fish species, are known to carry thiaminase, an enzyme known to degrade or destroy the thiamine in the gut content of consumers (Ishihara et al. 1973). The accumulation of thiaminase due to the over-reliance on anchovies as a food source is likely depleting the available thiamine in the breeding female salmon, and these low concentrations of thiamine are then being transferred into the eggs (Salmon Life History Team 2021). The fry hatch with depleted thiamine levels, and TDC contributes to their increased mortality during their development stages (Honeyfield et al. 2005).

Using data from 1997 through 2018, a fecal analysis study of nursing female sea lion pups at San Miguel Island off the coast of Los Angeles, California revealed sharp reductions in the occurrence of Pacific sardine, northern anchovy, and Pacific mackerel. While Pacific sardine and Pacific mackerel have exhibited gradual gains in fecal occurrence in recent years, northern anchovies rebounded in 2015 to levels exceeding their occurrence prior to 2008. Northern anchovy occurrences have remained at those levels in subsequent years. Conversely, market squid and Pacific hake have displayed marked reductions in occurrence since 2014 and 2015 respectively (Thompson et al. 2019). This shift in prey diversity and abundance was also observed further north in a gut content analysis of Chinook salmon captured in fisheries off California's central coast in 2020-2022 (Mantua, personal communication). The gut content study revealed that northern anchovies were responsible for 97% of gut contents by weight in Chinook salmon sampled in ocean fisheries off the Central California coast in summer 2020 (Mantua et al. 2021). Both studies point towards a reduction in prey diversity which could be resulting in Chinook salmon's increased intake of anchovies and thiaminase, impairing their ability to maintain sufficient thiamine concentrations.

A recent study of California's Central Valley winter-run Chinook salmon determined that pre-spawn females that received thiamine injections produced eggs with higher egg thiamine concentrations and increased viability of fry. Using the results of lab studies where treated and untreated eggs were hatched and juveniles reared in a controlled environment, a model was developed establishing threshold thiamine concentrations in eggs for viability of fry. The model predicts 95% fry viability for egg thiamine concentrations exceeding 5 nmol/g and near 100% viability for egg thiamine concentrations exceeding 8 nmol/g (Bell 2020). This model was then used to assess the prevalence of thiamine deficiency in Chinook salmon eggs from California hatchery sites. In 2020 late fall-run Chinook salmon at the Coleman National Fish Hatchery and winter-run Chinook salmon at the Livingston Stone National Fish Hatchery, nearly 50% of sampled eggs contained total thiamine concentrations below 5 nmol/g (Southwest Fisheries Science Center 2021). Due to the proportion of sampled eggs that had concentrations below 5 nmol/g, it was necessary to assess egg thiamine concentrations in other California river systems such as the Klamath River Basin (KRB) to see if it was experiencing similar results.

In 2023, demolition is scheduled to begin as part of a project to remove four dams on the Klamath River. The scope of the work comprises the largest dam removal project in world history. Hundreds of miles of spawning ground will be restored to the anadromous fish of the Klamath River for the first time in over a century. As a result, it is critical to understand the mechanisms that drive TDC, any contributing variables, and consider natural solutions (and management options more generally) to the issue that may be reinforced through conservation policy and stewardship of the river. The report summarizes egg thiamine concentrations for KRB Chinook salmon populations sampled in 2020 and 2021. It will also examine potential relationships between water temperature and distance of spawning migration to egg thiamine concentration, and it will also assess the relationships between egg diameter and egg thiamine concentration.

Methods

Chinook salmon egg samples were collected at three distinct locations within the KRB: the estuary of the Klamath River, Iron Gate Hatchery, and Trinity River Hatchery, representing variations in water quality, salinity, and temperature, migration routes and distance, habitat (freshwater vs. brackish), and time of collection. Samples consisted of unfertilized eggs collected from spawning and pre-spawning females. Once collected, egg samples were transported on dry ice, stored in -80 ^{o}C freezers, and shipped in dry ice to Dr. J. Rinchard (SUNY Brockport) where all the thiamine content analyses and egg diameter measurements were completed. One gram of eggs were used to extract free thiamine (TH), thiamine monophosphate (TMP), and thiamine pyrophosphate (TPP). After extraction, concentrations were determined with a high-performance liquid chromatograph (HPLC) system, and individual concentrations of the above vitamers were combined to determine egg thiamine concentrations. Each egg sample was analyzed via the HPLC system twice. For instances where the HPLC system analysis resulted in two distinct egg thiamine concentrations for an individual sample, the mean was taken of the two egg thiamine concentrations and assigned as the representative egg thiamine concentration for that sample.

As part of his ongoing master's thesis project, Jarrod Ludwig (SUNY Brockport) recorded egg diameter measurements. Per fish, a maximum of ten eggs (when available) were selected. Selected eggs were placed in a petri dish under a dissecting microscope and measured via Celestron Digital Microscope Imager HD software. The software system was calibrated to one millimeter, and each egg was measured from the outside of the egg shell directly through the center to the opposing outside of the egg shell. Only unbroken and the most spherical eggs were selected for analysis. The median measured diameter was calculated for each fish and assigned as the representative egg diameter for that fish.

Using Microsoft Excel (2019 MSO, Version 2304 Build 16.0.16327.20200, 64-bit), results of both egg thiamine concentration and egg diameter were organized into sample groups based on the location and date sampled. A single sample group was created for fish sampled in the Klamath River estuary in 2021 only (*Lower Klamath 2021*). Two years of data were collected at Iron Gate Hatchery and Trinity River Hatchery (*Iron Gate Hatchery 2020, Iron Gate Hatchery 2021, Trinity River Hatchery 2020, and Trinity River Hatchery 2021*). Additionally, analysis was performed at the sample site location and full basin level, identified as *Iron Gate Hatchery, Trinity River Hatchery*, and *Klamath River Basin* sample groups.

Publicly available mean daily water temperature data (DegC) was acquired via the United States Geological Survey's water data site (https://waterdata.usgs.gov) from gage stations at three distinct locations within the KRB, representing variations in migration routes and distances: Orleans, Klamath R NR Klamath, and Trinity R at Hoopa from August 1st through October 31st in 2020 and 2021. The time span of water temperatures and data years were selected to correspond with the fish sampling dates and the time of year that the sampled fish were most likely migrating through the KRB, based on the date they were sampled at the Iron Gate and Trinity River hatcheries. R Studio software (version 4.2.1, 2022-06-23 ucrt) was used to create scatter and box plots to depict variations in location and recorded temperatures from year to

year. R studio was also used to perform simple linear regression analysis to test predictability of median egg diameter on egg thiamine concentration.

Results

Lower Klamath 2021 Sample Group

The 43 Chinook salmon, all sampled in 2021 near the mouth of Klamath River, displayed a median egg thiamine concentration, expressed in nmol/g, of 19.8 (Range = 8.8-28.2; IQR = 16.0-22.5; Figure 1). This sample group was also assessed based on the date they were sampled. Fish sampled in August are identified as "Early." Fish sampled in September are identified as "Middle," and fish sampled in October are identified as "Late." The Early sample group, consisting of 20 sampled fish, displayed a median egg thiamine concentration of 21.7 nmol/g (Range = 8.8-25.2; IQR = 17.1-23.5). The Middle sample group, consisting of 20 sampled fish, displayed a median egg thiamine concentration of 17.5 nmol/g (Range = 11.7-28.2; IQR = 14.7-20.4), and the Late sample group, consisting of 3 sampled fish, displayed a median egg thiamine concentration of 21.3 nmol/g (Range = 17.1-22.2; IQR = 19.2-21.8).

Since these fish were sampled in close proximity to the mouth of the river, the distance of migration for this sample group is considered to be 0 kilometers. Water temperature data was not reviewed for this sample group due to the sampling location's interaction with the ocean through tide cycles.

Trinity River Hatchery 2020 Sample Group

This sample group included the 60 Chinook salmon sampled at Trinity River Hatchery in 2020 (30 spring-run; 30 fall-run). The spring run-fish displayed a median egg thiamine concentration of 12.9 nmol/g (Range = 8.0-18.2; IQR = 11.6-14.6), while the fall-run fish displayed a median egg thiamine concentration of 16.2 nmol/g (Range = 8.5-23.8; IQR = 13.4-19.1; Figure 2).

The fitted regression model for the fall-run group was: Egg thiamine concentration = 24.0 – 1.1(median egg diameter). The overall regression was not statistically significant (R^2 = 0.007, F(1, 28) = 0.200, p = 0.658). It was found that median egg diameter did not significantly predict egg thiamine concentration (β = -1.1, p = 0.658).

Trinity River Hatchery 2021 Sample Group

This sample group included the 61 Chinook salmon sampled at Trinity River Hatchery in 2021 (31 spring-run; 30 fall-run). The spring-run fish displayed a median egg thiamine concentration of 14.4 nmol/g (Range = 9.3-20.9; IQR = 13.1-15.7), while the fall-run fish displayed a median egg thiamine concentration of 11.6 nmol/g (Range = 6.3-18.3; IQR = 10.8-14.0; Figure 2).

Iron Gate Hatchery 2020 Sample Group

This sample group included the 30 fall-run Chinook salmon sampled at Iron Gate Hatchery in 2020. The fish in this group displayed a median egg thiamine concentration of 19.1 nmol/g (Range = 11.5-30.5; IQR = 16.4-21.7; Figure 1).

Iron Gate Hatchery 2021 Sample Group

This sample group included the 30 fall-run Chinook salmon sampled at Iron Gate Hatchery in 2021. The fish in this group displayed a median egg thiamine concentration of 12.1 nmol/g (Range = 5.5-20.5; IQR = 9.7-14.8; Figure 1).

Basin-Scale Analysis for 2021 Samples

All 134 Chinook salmon sampled during 2021 displayed a median egg thiamine concentration of 14.3 nmol/g (Range = 5.5-28.2; IQR = 11.7-18.2; Figure 3).

Simple linear regression was used to test if median egg diameter significantly predicted egg thiamine concentration for the 2021 samples when samples were available from all sites in the same spawning year (Lower Klamath, Iron Gate Hatchery, and Trinity River Hatchery; Figure 4). The fitted regression model was: Egg thiamine concentration = 36.4 – 3.1(median egg diameter). The overall regression was statistically significant (R^2 = 0.258, F(1, 132) = 45.970, p <0.000). It was found that median egg diameter did significantly predict egg thiamine concentration (β = -3.1, $p < 0.000$).

Water Temperature

Mean daily Aug 1-Oct 31 water temperature, all expressed in degrees Celsius, ranged from 10.6-24.4 with a median of 18.6 at Orleans gage station in 2020 (10.9-25.3, median of 18.8 in 2021), 11.8-23.7 with a median of 19.2 at R NR Klamath gage station in 2020 (11.7-24.2, median of 19.0 in 2021), and 10.5-24.3 with a median of 17.9 at Trinity R Hoopa gage station in 2020 (11.6-25.1, median of 18.0 in 2021; Figure 5).

Discussion

Egg Thiamine Concentration Comparison and Relationship to Distance of Migration and Water Temperature

Sample groups for the purposes of egg thiamine concentration comparison were identified based on the location of the sample, time of year collected (Trinity River Hatchery sample groups only), and the year in which they were sampled (Figure 1). The Lower Klamath 2021 sample group displayed the highest median egg thiamine concentration at 19.8 nmol/g. This sample group displayed the highest upper limit of its IQR at 22.5 nmol/g. Other variables that were considered in this study in terms of how they potentially contribute to egg thiamine concentration included distance of migration and water temperature. We hypothesized that distance of migration would be a factor in depleted egg thiamine concentrations based on the

effort expelled by the fish to reach the sample location as well as the total time spent removed from the ocean environment where the thiamine had been accumulated in the fish's body. It was also hypothesized that exposure to warmer water temperatures for longer durations would also deplete egg thiamine concentrations.

Based on the Lower Klamath location's proximity to the ocean, this sample group was expected to display the highest median egg thiamine concentration. The fish sampled in this group had not yet endured a migration to spawning grounds upriver, so the effort exerted to traverse the river prior to sampling was far less than the other sample groups. Additionally, this sample location is still heavily influenced by the ocean ecosystem through influxes of water at ocean temperature as well as ocean nutrients through daily tidal cycles. If sample location water temperature and distance of migration are factors that contribute to depleted levels of egg thiamine concentration, this sample group would be least impacted by differences in river water temperature as compared to ocean temperature and distance of migration. Additionally, eggs sampled in the lower river may have been immature, a likelihood supported by the fact that egg diameter was consistently smaller at this site than at the upriver hatchery sites.

Trinity River Hatchery sample groups represented a migration distance of 247 river kilometers. The 2021 Trinity River Hatchery Spring-Run sample group displayed a median egg thiamine concentration of 14.4 nmol/g and IQR upper limit of 15.7 nmol/g, and the 2021 Trinity River Hatchery Fall-Run sample group displayed a median egg thiamine concentration of 11.5 nmol/g with an upper IQR limit of 14.0 nmol/g. Iron Gate Hatchery sample groups migrated 305 river kilometers prior to being sampled, and the 2021 Iron Gate Hatchery sample group displayed a median egg thiamine concentration of 12.1 nmol/g with an IQR upper limit of 14.8 nmol/g. The 2021 sample groups did display a distance effect on egg thiamine concentrations. Except for Trinity River Hatchery's fall-run 2021 sample group displaying slightly lower concentrations than the 2021 sample group from Iron Gate Hatchery, the sample groups with the shortest migrations displayed the highest egg thiamine concentrations (Figure 6).

2020 sample groups did not display the same distance effect, but data was only available to compare the Trinity River Hatchery sample groups to the Iron Gate Hatchery sample group. Data for the Lower Klamath was not collected in 2020. The 2020 Trinity River Hatchery Spring-Run sample group displayed a median egg thiamine concentration of 12.9 nmol/g and IQR upper limit of 14.6 nmol/g, and the 2020 Trinity River Hatchery Fall-Run sample group displayed a median egg thiamine concentration of 16.2 nmol/g with an upper IQR limit of 19.1 nmol/g. Iron Gate Hatchery's 2020 sample group displayed a median egg thiamine concentration of 19.1 nmol/g and an IQR upper limit of 21.65 nmol/g (Figure 6). While the Iron Gate sample group displayed higher egg thiamine concentrations despite the longer migration distance, the Iron Gate Hatchery 2020 sample group did appear to be out of pattern when compared to all other sample groups covering the two years of data. Additional environmental factors such as ocean prey availability and diversity, weather patterns, stream flow, and local water temperatures near Iron Gate Hatchery could have contributed to the measurements observed in 2020.

Water temperature data were also reviewed as a potential contributor to differences in egg thiamine concentration between the 2020 and 2021 sample groups at Iron Gate Hatchery. Fish sampled at Iron Gate Hatchery in both years would have migrated past the Orleans stream temperature gauge station. However, the range of temperatures experienced at the station between 1 August and 31 October of each year were comparable. The median mean daily water temperature at the station in 2021 was 18.8 °C, and the median mean daily water temperature at the station in 2020 was 18.6 °C. The upper limit of the IQR and maximum mean daily temperature recorded in 2021 were 20.9 °C and 25.3 °C respectively, and the upper limit of the IQR and maximum mean daily temperature recorded in 2020 were 22.9 °C and 24.4 °C respectively.

Similar year-over-year differences in mean daily water temperature were observed at the Trinity River Hoopa water gage station, which is the station that the Trinity River Hatchery sample groups passed through during their migration. Based on data collected from August 1, 2020 through October 31, 2020, the Trinity Hoopa water gage station displayed a median mean daily water temperature of 17.9 °C with an upper quartile limit of 22.4 °C, and the station displayed a median mean daily water temperature of 18.0 °C with an upper quartile limit of 19.8 °C over that same time span in 2021. Based on the egg thiamine concentration results from 2020 and 2021 at the Iron Gate and Trinity River Hatcheries combined with the mean daily water temperature results observed in the same study years at Orleans gage station and Trinity Hoopa gage station, a relationship between egg thiamine concentration and water temperature could not be identified.

Relationship Between Egg Thiamine Concentration and Median Egg Diameter

For each fish that was sampled for egg thiamine concentration in this study, egg diameter measurements were also recorded. We hypothesized that egg thiamine concentrations would decrease as egg diameter measurements increased. Since Chinook salmon stop feeding once they enter the river system, they would not be accumulating new stores of thiamine, and thiamine content could only decrease as they continue their migration. Therefore, we would expect samples from the Lower Klamath section of the river to have smaller, less-developed eggs with higher thiamine concentrations based on their smaller size and less time elapsed to deplete thiamine stores since the last feeding. Fish sampled at Trinity River Hatchery and Iron Gate Hatchery spent more time in the river allowing eggs to grow and develop further. We would expect to see larger egg diameter measurements but lower egg thiamine concentrations since these fish still have not taken in thiamine in their diet since their last ocean feeding and now have larger volume eggs requiring the same, or depleted, thiamine stores that the fish entered the mouth of the river with.

Simple linear regression analysis was performed at the basin-scale using only 2021 data. Data was not collected on the Lower Klamath in 2020, and by isolating 2021 data only, we were able to limit impacts from differences in year-to-year environmental variables such water temperature, stream flows, and ocean prey availability. The basin-scale 2021 data did display a significant relationship between median egg diameter and egg thiamine concentration. With a p

value < 0.000, the basin-scale analysis displays lower egg thiamine concentrations as median egg diameter increases. The Lower Klamath 2021 sample group recorded the highest median egg thiamine concentration at 19.8 nmol/g and the smallest median egg diameter at 6.1 mm. This group also represented the shortest migration at 0 kilometers. The Trinity River Hatchery sample groups from 2021 displayed median egg thiamine concentrations of 14.4 nmol/g in the spring and 11.6 nmol/g in the fall. Median egg diameters were 7.1 mm for both groups, and this group had migrated 247 kilometers prior to sample collection. Finally, the Iron Gate Hatchery 2021 sample group recorded a median egg thiamine concentration of 12.1 nmol/g and a median egg diameter of 7.6 mm, representing 305 kilometers of migration. As eggs have more time to grow and develop along longer migration routes, egg thiamine concentration decreases in the larger-sized eggs (Figure 4).

Recommendations

Future studies should work to obtain data on variables specific to each sample fish to allow for stronger correlation and predictability analyses. In the case of assessing the relationship between egg diameter and egg thiamine concentration, the egg diameter recorded should be that of the same egg being sampled for egg thiamine concentration creating a direct relationship. Using a measure of central tendency from a collection of egg samples as representative for the egg thiamine concentration introduces variance within the egg diameters of an individual fish, and those individual eggs could also have variance in their egg thiamine concentration. Only introducing egg diameter measurements from eggs that were also sampled for egg thiamine concentration would eliminate the variance introduced by representative egg diameter measurements from eggs that were never measured for egg thiamine concentration. Specific variable measurements for individual samples will also facilitate multivariate analysis.

Water temperature is a particularly difficult variable to assess when attempting to establish strength of relationship or predictability and is best used for observational comparison only. Currently, we are not able to definitively determine any individual fish's exposure to changes in water temperature during its spawning migration, and we do not know the length of time that any individual fish has been in the river system since exiting the ocean environment. In the KRB, there can be great variability observed in the water temperature based on weather, canopy cover, fish access to cold water tributaries, and flow rates managed by upstream dams among other variables. A recommended metric which may provide additional insight into an individual fish's water temperature experience in the river system would be the number of days specified locations in the river system exceed various threshold temperatures during the spawn migration time period. This would also require recording the date each sampled fish was removed from the river system. Daily water temperatures can also be recorded at the hatchery sites to determine if there is a relationship between water temperature and the viability of fry.

It is also recommended that egg thiamine concentration data baselines in the KRB be established for future comparative analyses. Later in 2023, the KRB will undergo the largest dam removal project in world history drastically changing the river ecosystem and returning it to a more-wild status. Establishing a baseline of data prior to dam removal will allow for analysis

post dam removal to determine if the removal of the dams is having the anticipated effects. The baseline can also be applied to other future natural and anthropogenic events such as individual extreme weather events, climate change, wildfires, and changes in agricultural water allocations to name a few. Continued measurement in addition to these established baselines in conjunction with standardized data collection, analysis, and reporting procedures in other salmonid rivers along the Pacific west coast can inform expected impacts from such events in other river systems and potentially provide opportunities for mitigation to minimize impacts to salmonid populations.

Conclusion

Out of 224 Chinook salmon egg thiamine concentration samples taken from the KRB in 2020 and 2021, no eggs exhibited an egg thiamine concentration below the 95% viability threshold of 5 nmol/g. Nine total samples (five from Iron Gate Hatchery, four from Trinity River Hatchery) displayed egg thiamine concentrations below the 100% viability threshold of 8 nmol/g, representing only 6.9% of the total population sampled. These results are encouraging for the Chinook salmon population within the KRB when compared to results displayed from Chinook salmon hatcheries in California's Central Valley, two of which displayed results from samples in 2020 with nearly 50% of the Chinook salmon population exhibiting egg thiamine concentrations less than 5 nmol/g.

However, data prior to 2020 is not available for the KRB, and the results of this study are not able to identify how the results from 2020 and 2021 compare to historical egg thiamine concentration levels. Continued data compilation and analysis are necessary to monitor shifts in egg thiamine concentrations of future Chinook salmon runs within the basin. Additional data and analysis on relationships between water temperature, distance of migration, and egg diameter are also needed to build a larger data set that can more accurately depict the strength of the relationships between these variables and egg thiamine concentration. With the basin set to undergo the largest dam removal project in world history in 2023, a baseline of egg thiamine concentration data will be necessary to compare to measured impacts once the dams have been removed and the river system has been returned to a more-wild state. The KRB will serve as a benchmark for future dam removal proposals in other river systems, and the ability to assess post-dam removal impacts will be crucial to properly inform those proposals.

References

Bell, H. N. 2022. Assessment of the Effects of Thiamine Deficiency on the Survival, Physiology, and Behavior of Early Life-Stage Winter-Run Chinook Salmon. *UC Davis*. ProQuest ID: Bell_ucdavis_0029M_21136. Merritt ID: ark:/13030/m5sg04ft. Retrieved from https://escholarship.org/uc/item/465026qq

Harder, AM., et al. 2018. Thiamine deficiency in fishes: causes, consequences, and potential solutions. Rev Fish Biol Fisheries. 28: 865-886.

Honeyfield, D.C., J.P. Hinterkopf, J.D. Fitzsimons, D.E. Tillitt, J.L. Zajicek, and S.B. Brown 2005. Development of thiamine deficiencies and early mortality syndrome in lake trout by feeding experimental and feral fish diets containing thiaminase. Journal of Aquatic Animal Health. 17: 4–12.

Ishihara, T., M. Yasuda, and H. Morook 1972. Studies on thiaminase I in marine fish – I. Thiaminase in anchovy. Bull. Jap. Soc. Sci. Fish. 38: 1281-1287.

Mantua, N. et al. 2021. Mechanisms, Impacts, and Mitigation for Thiamine Deficiency and Early Life Stage Mortality in California's Central Valley Chinook Salmon. North Pacific Anadromous Fish Commission Technical Report. 17: 92-93.

Reed, A.N., F.E. Rowland, J.A. Krajcik, and D.E. Tillitt 2023. Thiamine Supplementation Improves Survival and Body Condition of Hatchery-Reared Steelhead (Oncorhynchus mykiss) in Oregon. Vet Sci. 10(2): 156.

Riley, S.C., and A.N. Evans 2018. Phylogenetic and ecological characteristics associated with thiaminase activity in Laurentian Great Lakes fishes. Trans Am Fish Soc. 137:147–157

Salmon Life History Team. 2021. Monitoring Thiamine Deficiency in California Salmon. NOAA Fisheries. Retrieved from http://fisheries.noaa.gov/west-coast/science-data/monitoring-thiamine- deficiency-california-salmon.

Southwest Fisheries Science Center 2021. Thiamine Deficiency in West Coast Salmon. NMFS SWFSC Report (March 2021). 1-4. Retrieved from pcouncil.org/documents/2021/02/e-1-attachment-1-1thiamine-deficiency-in-west-coast-salmon.pdf/.

Thompson, A., I.D. Schroeder, S. Bograd, and E.L. Hazen. 2019. State of the California Current 2018–19: a novel anchovy regime and a new marine heatwave? Calif. Coop. Ocean. Fish. Invest. Rep. 60: 1–65.

Figures

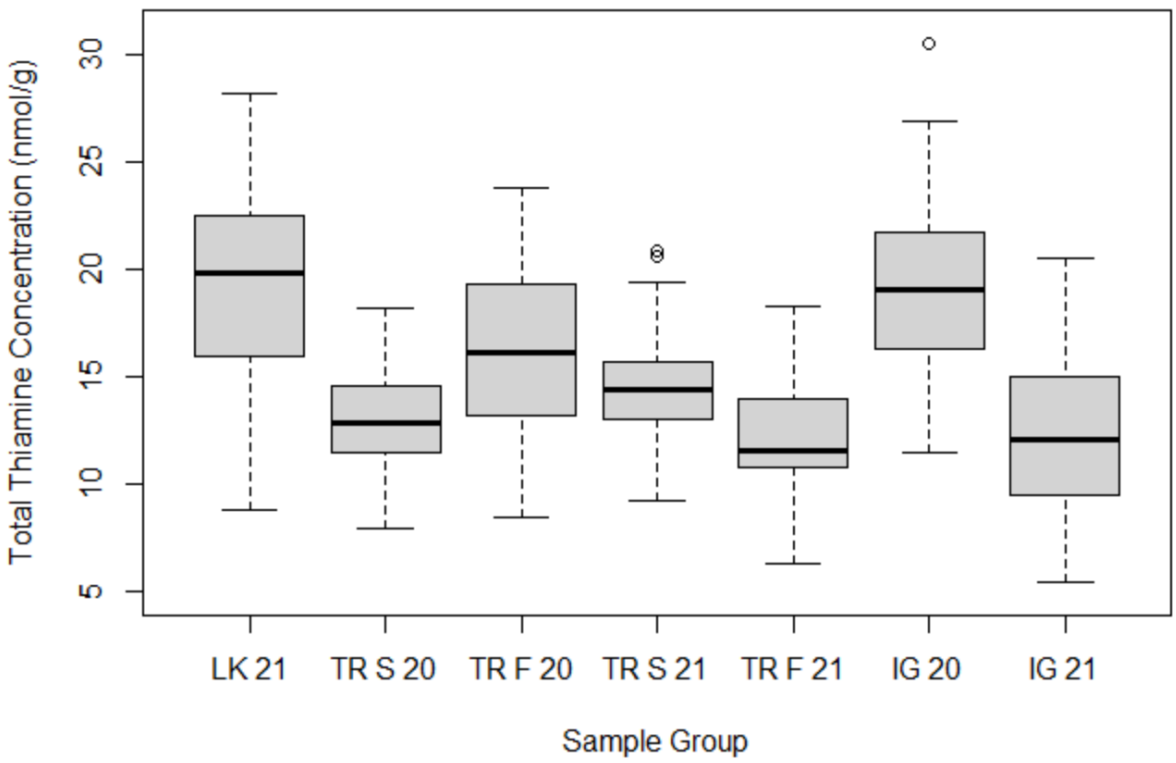

Figure 1. Box plot depicting total egg thiamine concentrations of each of the five sample groups included in this study; "LK 21" = Lower Klamath 2021, "TR S 20" = Trinity River Hatchery Spring 2020, "TR F 20" = Trinity River Hatchery Fall 2020, "TR S 21" = Trinity River Hatchery Spring 2021, "TR F 21" = Trinity River Hatchery Fall 2021, "IG 20" = Iron Gate Hatchery 2020, "IG 21" = Iron Gate Hatchery 2021

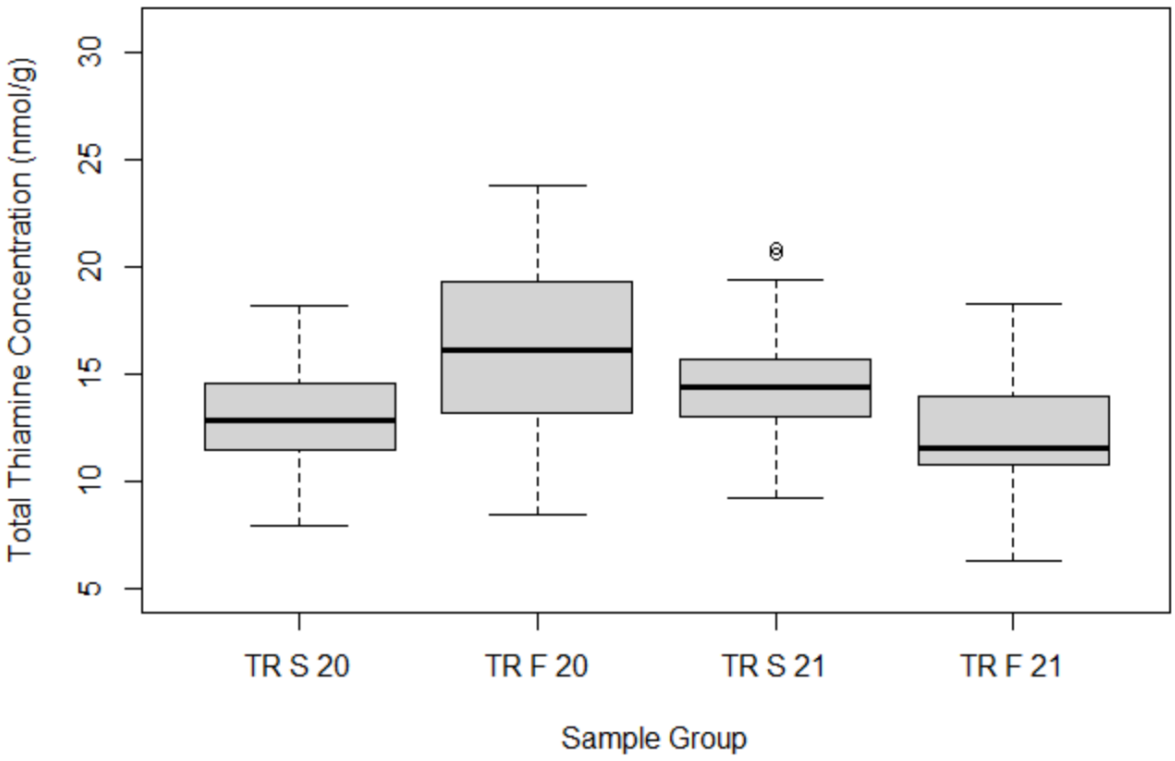

Figure 2. Trinity River Hatchery – Spring and Fall-run egg thiamine concentrations for 2020 and 2021

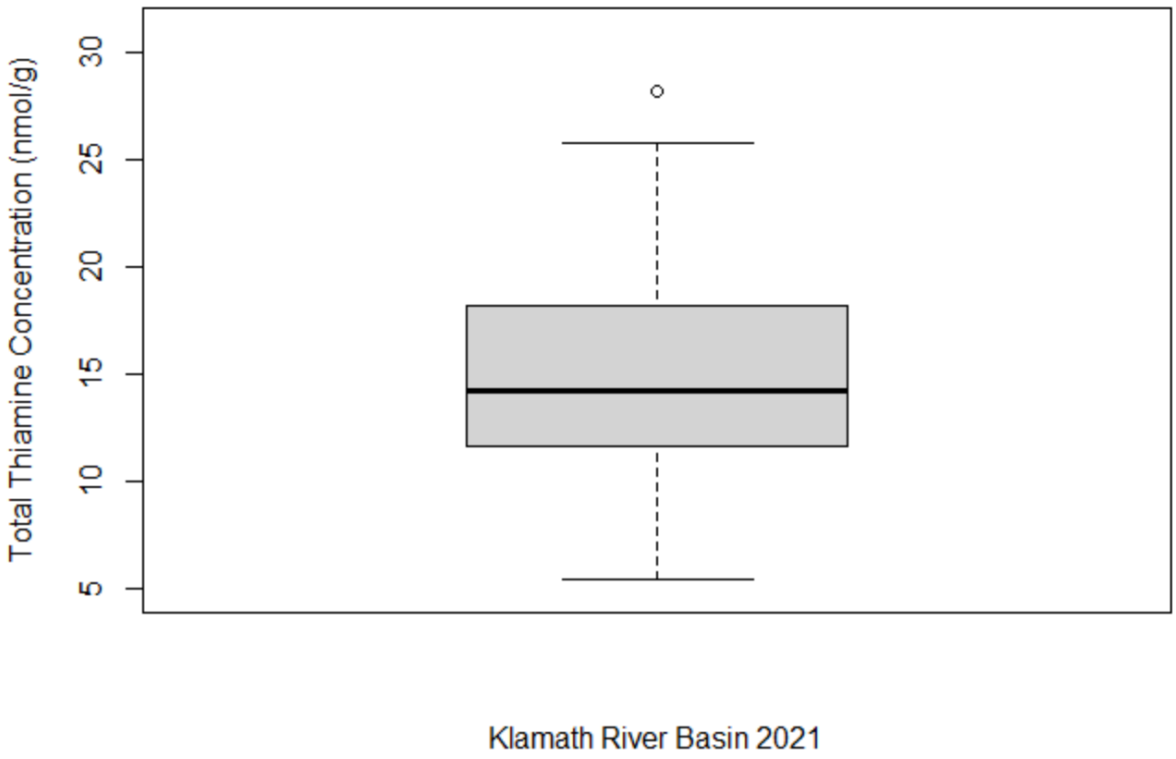

Figure 3. Egg thiamine concentration of the Klamath River Basin in 2021

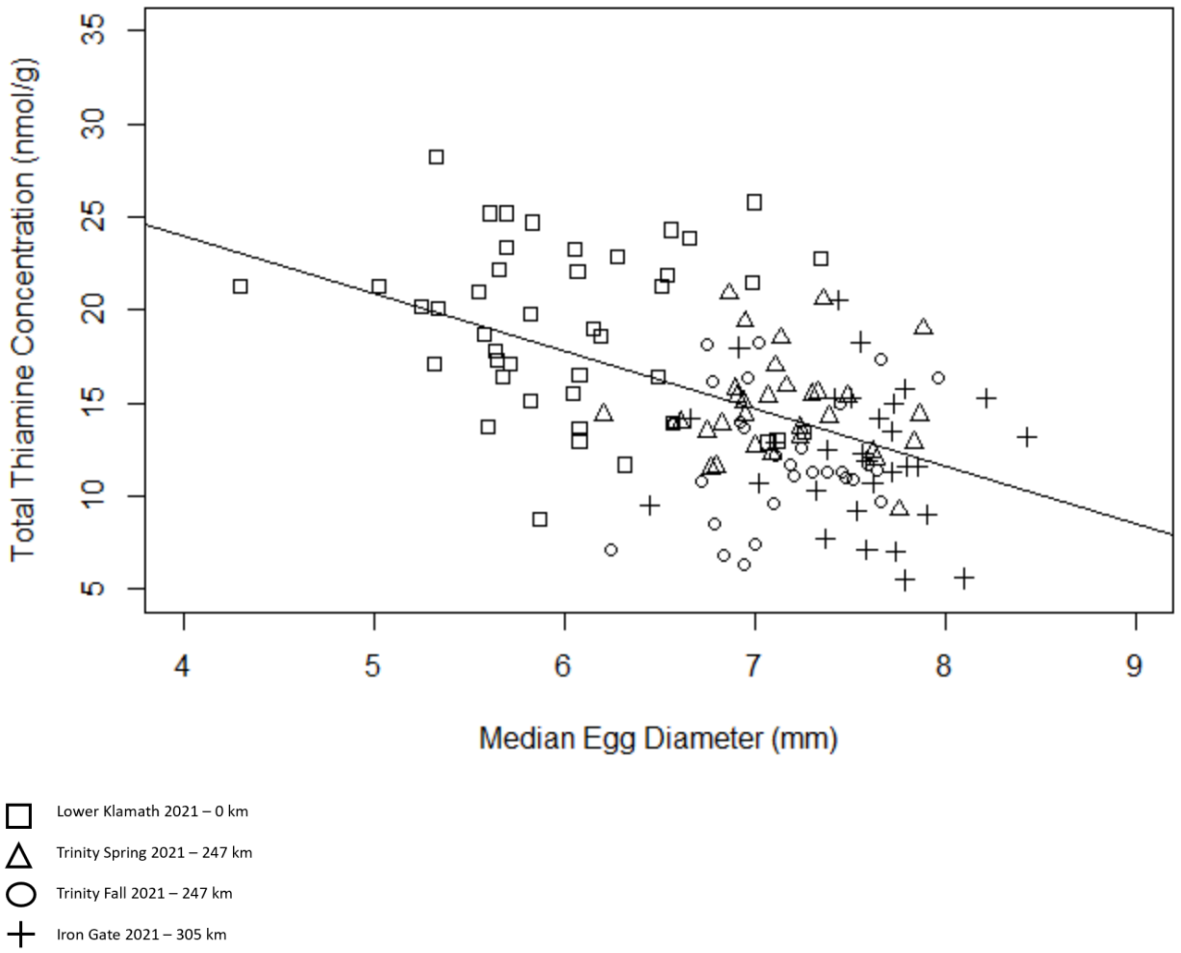

Figure 4. Relationship between median egg diameter and egg thiamine concentration while denoting differences in sample location and migration distance of sampled fish

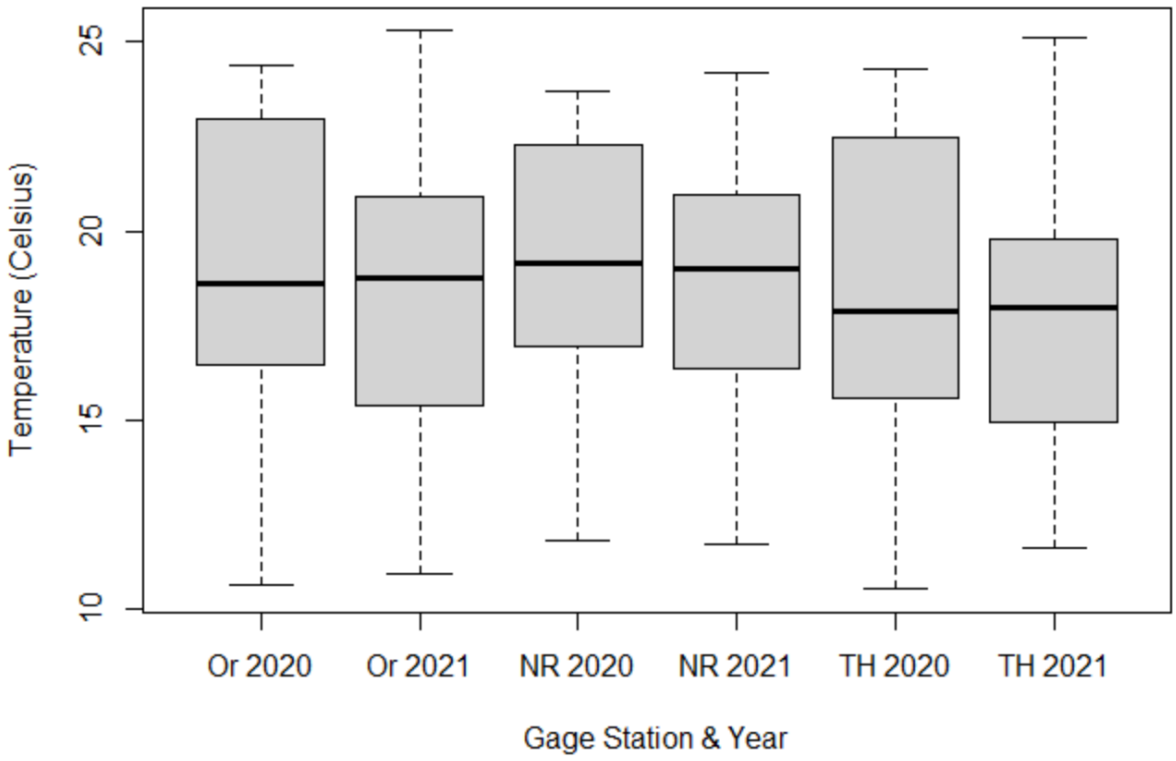

Figure 5. Daily mean water temperatures were recorded from August 1st to October 31st in years 2020 and 2021; "OR" = Orleans gage station, "NR" = Klamath R NR gage station, "TH" = Trinity Hoopa gage station

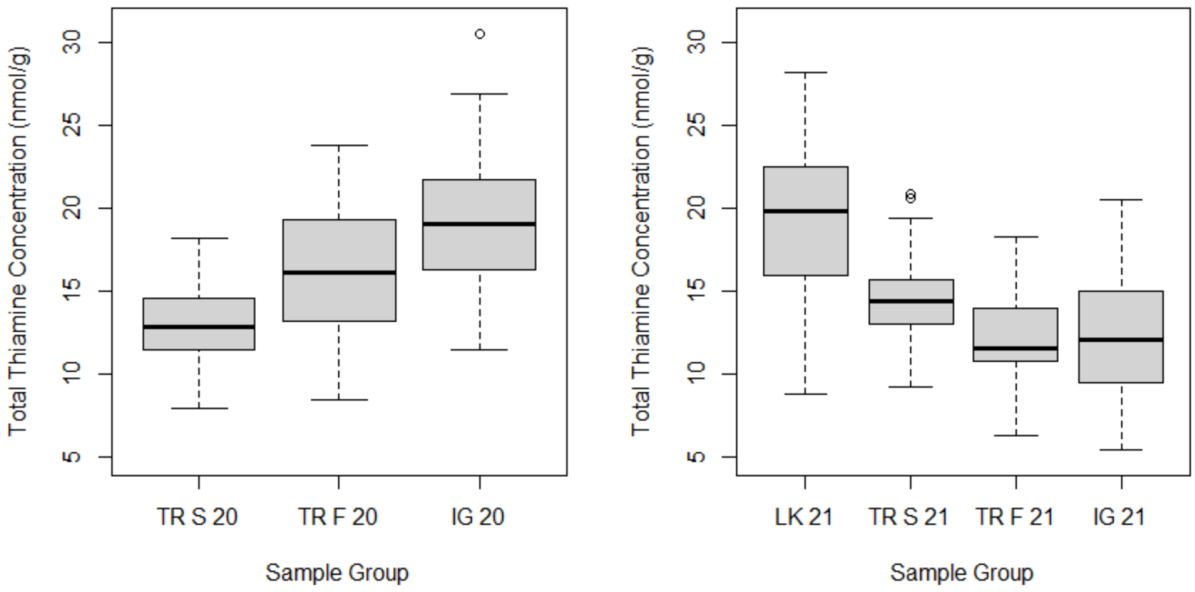

Figure 6. Egg thiamine concentration of all sample groups separated by year