APPENDIX D. DISEASE HISTORY AND PATHOGEN RISKS AT WARM SPRINGS NATIONAL FISH HATCHERY¹

I. Fish Health Issues

Warm Springs National Fish Hatchery (NFH) faces many fish health challenges associated with increasing summer air and water temperatures, a surface water source containing adult salmon, and increased frequency of summer droughts. For example, external parasitic infections of *Ichthyophthirius multifiliis* (Ich) and bacterial infections of *Flavobacterium columnare* (Columnaris) require frequent treatments with formalin and Chloramine-T, respectively, to avoid significant disease losses. Warm water disease problems at Warm Springs NFH have sometimes been severe enough to warrant relocation of Spring Chinook Salmon (*Oncorhynchus tshawytscha*) subyearlings and adults to Little White Salmon NFH where water temperatures are much cooler during the summer months. Disease risk is also higher in the weeks and months after summer temperatures have subsided because accumulated stress from warm water temperatures has long-term effects on disease resistance. These long term stress responses may have contributed to three outbreaks of Bacterial Kidney Disease (BKD) that occurred between 2015 and 2019 in Spring Chinook Salmon just prior to release into the Warm Springs River in April.

Warm Springs NFH must use antibiotics often to control bacterial infections in juvenile Chinook Salmon during the summer months (June – September) when average water temperatures are greater than 15 °C (60 °F) and often approach 21 °C (70 °F). These warm water temperatures, coupled with a surface water supply known to contain many different fish pathogens, trigger disease outbreaks that, if left untreated, would result in the loss of the majority of the fish on station. Since 2012, annual treatment of Chinook salmon juveniles with Teramycin-200 or Aquaflor has been necessary to control Columnaris and Motile Aeromonas Septicemia (MAS). Hatchery management and fish health staffs do not believe the reliance on drugs and chemicals is sustainable or a best management practice (BMP).

In addition to increased incidence of infectious diseases, warmer water temperatures at Warm Springs NFH also contribute to husbandry, rearing, and behavioral issues. In recent years, adult hatchery broodstock have faced increasingly non-optimal migration temperatures (>16 °C or >60 °F) that affect spawning behavior and impact developing eggs causing uneven egg maturation, lower egg eye-up, developmental issues at hatch, and high early-rearing "drop-out" (cessation of feeding in competition with other fish). Those issues also contribute to physiological stress as water temperatures increase during the spring and summer.

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II. Primary concerns and current treatment/mitigation measures

The three pathogens of greatest concern during the summer months are (a) the bacterium that causes Columnaris (*Flavobacterium columnare*), (b) the parasite that causes Ich (*Ichthyophthirius multifiliis*), and (c) the parasite *Ceratonova shasta*. These pathogens often co-infect Chinook Salmon at Warm Springs NFH making treatment complicated with several chemicals and drugs being applied concurrently.

In addition, *Ichthyobodo* spp., the causative agent of Ichthyobodosis (costia disease, not to be confused with "Ich"), has been a cause of significant acute mortality during early rearing in the nursery building if temperatures reach 13 °C (55 °F) or greater.

A. Flavobacterium columnare (F. col.)

This bacterium is ubiquitous in the aquatic environment and causes Columnaris disease. Since 2014, Columnaris has caused significant mortality among juvenile Chinook Salmon at Warm Springs NFH. In addition, adults trapped for broodstock returning to Warm Springs NFH have often had a significant external infection with *F. col.* Survival and growth of this bacterium is temperature and water-chemistry dependent. Disease is likely to occur when water temperature is above 16 °C (60 °F). Increased water hardness, organic matter, and a low-level salinity provide an environment favorable for this bacterium. This disease could become more severe in the summer months and more prevalent into spring and fall in response to projected climate changes.

Current measures to address F. col.:

- Water flow and turnover rates have been maximized within the physical constraints of the hatchery and watershed by (a) increasing the amount of water flow into the rearing units or (b) reducing the rearing volume during the summer months, thereby increasing water turnover rates.
- The density index is maintained at or below the general recommendation of 0.20 lbs./ft³/inch throughout the rearing cycle, or one adult broodfish per gallon per minute (gpm).
- Broodstock are held at $10 \text{ °C} \pm 1 \text{ °C}$ (50 °F $\pm 2 \text{ °F}$) to reduce the growth rate of both *F*. *col.* and Ich, and to reduce pre-spawn mortality of adults due to the parasite *C. shasta*.
- Low energy feeds with a lower protein and lipid content are used during summer when water temperatures are high to decrease metabolic loads and stress.
- Frequent and regular monitoring has greatly improved the ability of hatchery staff to deliver Chloramine-T early in the external infection process, thus increasing the likelihood of controlling the infestation and reducing mortalities.
- Adult broodfish initially receive Chloramine-T two to three times per week to reduce the *F. col.* load when broodstock are first collected.
- Based on Columnaris disease history, Terramycin-200 has reduced incidence of systemic infection due to *F. col.* when used prophylactically prior to infection.

B. Ichthyophthirius multifiliis (Ich)

This parasite causes White Spot Disease (aka, Ich). Adult Chinook Salmon trapped for broodstock at Warm Springs NFH have a significant Ich infection. The life cycle of Ich is temperature dependent; the parasite is able to complete its life cycle faster as water temperatures increase from 12 °C (54 °F) to 22 °C (72 °F) (Table B2, Appendix B). If untreated, thousands of infective parasites are formed and released into the environment with the completion of each new life cycle. One way of combating Ich is to increase water flow or decrease fish density (or increase water volume), thus decreasing the encounter probability between the parasite and host. Unfortunately, this latter strategy is difficult, if not impossible, during periods of low water availability (e.g., during the summer months when water temperatures favor Ich). In addition, the immune system of salmonid fishes is significantly less effective at combatting Ich at the warmer temperatures that favor the parasite.

Current measures to address Ich:

- Water flow, turnover rates, density index, and temperature control, as described for preventing *F. col.*, are also beneficial to reduce the severity of Ich infections.
- Frequent and regular monitoring has greatly improved the ability of hatchery staff to deliver formalin early in the infection, thus increasing the likelihood of controlling infestation and reducing mortality. To control infection of Ich and prevent pre-spawn mortality, returning adults held at Warm Spring NFH are treated with formalin 3 5 days per week throughout the collection and maintenance cycle until spawning.
- For juvenile Chinook Salmon, use of an extra-label prescription for a low-dose, longerduration formalin treatment has led to better success treating the fish than results obtained previously with the approved labeled dose.

C. Ceratonova shasta (C. shasta)

This parasite is a microscopic multicellular myxozoan, currently poses two risks for hatchery and natural-origin Chinook Salmon in the Warm Springs River: (1) natural origin adults passed upstream of Warm Springs NFH, clinically infected with *C. shasta*, will likely die before spawning (pre-spawn mortality), and (2) pre-spawn mortality among infected adults retained for broodstock is elevated. Two potential future risks are (1) disease among natural origin juveniles in the Warm Springs River, and (2) infection of juvenile hatchery-origin Chinook Salmon at Warm Springs NFH.

C. shasta has a complex life cycle, and at Warm Springs NFH, is a problem only in adult fish as they return to the hatchery. The infectious lifecycle of *C. shasta* begins when the intermediate host, a polychaete worm, sheds *C. shasta* actinospores that infect a living fish or carcass. The parasite grows and reproduces, resulting in clinical disease and potential death of an infected fish. Mature parasites in a sick or dead salmonid fish (the primary host) shed myxospores that infect the polychaete worm (the intermediate host), thus completing the life

cycle. Fish-to-fish transmission of *C. shasta* does not occur. The polychaete worm can survive water temperatures greater than 24 $^{\circ}$ C (75 $^{\circ}$ F).

The parasite and polychaete intermediate host have been identified in the upper and lower Deschutes River Basin, but the polychaete worm does not appear to be present in the Warm Springs River upstream of the hatchery. Adults returning to the Warm Springs River carry and shed *C. shasta* myxospores, but the parasite cannot complete its life cycle and infect juvenile fish in the river or in the hatchery without the presence of the polychaete worm.

In general, *C. shasta* can cause disease when water temperatures reach 10 °C (50 °C) with the severity of mortality increasing as water temperature increases. An increase in water temperature coupled with a decrease in water flows of the Deschutes River would elevate *C. shasta* risk to upstream-migrating adults by increasing the likelihood of fish being exposed to the pathogen. The risk is further compounded by immunosuppression caused by decreased oxygen levels and increased temperatures. Higher future temperatures of the Warm Springs River would further increase the likelihood of clinical disease among upstream migrating adults prior to arriving at the hatchery.

Currently, *C. shasta* actinospores shed by infected polychaete hosts are encountered by both smolting Chinook Salmon during outmigration through the Deschutes and Columbia Rivers and by adults returning along the same route to spawn. Pre-spawn mortality due to *C. shasta* has been a problem with broodstock held at Warm Springs NFH when daily adult holding temperatures increase above 10 °C (50 °F). The number of natural-origin Spring Chinook Salmon returning to and spawning in the Warm Springs River, with river temperatures ranging from 13 - 21 °C (55 - 70 °C), has been decreasing for over a decade. If the polychaete intermediate host becomes established in the Warm Springs River, then the resulting infection of juvenile fish would most likely be devastating for both natural-origin and hatchery-origin Chinook Salmon.

Current measures to address *C. shasta*: Infection of *C. shasta* during migration cannot be prevented prior to adult fish arriving at the hatchery, and therapeutic treatments for infected fish are not available. At Warm Springs NFH, broodstock are held in chilled water to delay development of the parasite long enough for successful spawning to occur. Juvenile salmon are infected by the parasite in the Deschutes and Columbia rivers, but the impact of those infections on subsequent survival is unknown.

D. Conclusions

Since 2014, survival of Chinook Salmon at Warm Springs NFH has relied increasingly on the routine use of chemicals and antibiotics to minimize disease and mortalities. FWS staffs at Warm Springs NFH and the Columbia-Pacific Northwest Region Fish Health Program have concluded that the current situation is not sustainable over the long term, both in terms of fish health and human health and environmental safety. In addition to Columnaris and Ich, accumulated stress from non-optimal conditions (e.g., high water temperatures) and other

stressors (e.g., marking and tagging) have resulted in outbreaks of BKD among pre-smolt Chinook Salmon.

III. Other pathogens detected at Warm Springs NFH

Pathogens described in Section II above are currently the most obvious problems at Warm Springs NFH during the summer. However, infections by other parasites could play a significant physiological role reducing immunity during non-optimal summer conditions and, thus, contribute to the severity of Ich and Columnaris disease. These parasites (described below) are found in gills (*Nanophyetus* spp., *Sanguinicola* spp., *Echinochasmus* spp., *Trichodina* spp., *Saprolegnia* spp.), skin (*Nanophyetus* spp., *Saprolegnia* spp.), kidney (*Nanophyetus* spp., *Sanguinicola* spp., *Myxobolid* spp.), intestine (*Hexamita* spp.) and most internal organs (*Nanophyetus* spp.).

A. Nanophyetus spp.

This parasite is a fluke that has been identified at high levels in many organs of Chinook Salmon year-round at Warm Springs NFH, among both juveniles and adult broodstock. The outcomes and prevalence of this pathogen in response to higher water temperatures are unknown, but as water temperatures during the summer increase above 60 °F (16 °C), the immune systems of the fish are less likely to combat the parasite successfully. Higher temperatures may also increase the density of the snail host of this parasite.

B. Sanguinicola spp.

This parasite is a blood fluke that is detected at low numbers in the gills of Chinook Salmon in the early spring at Warm Springs NFH with an increasing load through the spring and summer. The parasite circulates in the blood and accumulates in internal organs, particularly the kidney. As the water temperature warms during June and July, the parasite emerges from the gills and causes damage to the secondary lamellae at a time when the environmental load of *F. col.* and Ich are increasing, and fish immunocompetency is compromised.

C. Echinochasmus spp.

This parasite is a fluke that is present in the gills within a month after fry are transferred from the indoor nursery tanks to outdoor raceways. This parasite is commonly found in gills of most fish species in Oregon. The osmoregulatory effects of this parasite to Spring Chinook Salmon under non-optimal conditions are unknown.

D. Trichodina spp.

This parasite is a single-celled ciliate that is detected in the spring or fall when water temperatures are fluctuating between optimal and non-optimal. Although this parasite is not a pathogen of primary concern, infection results in stress and compounds concurrent pathogenic infections.

E. Saprolegnia spp.

This parasite is a fungus-like oomycete is found secondarily in conjunction with Columnaris lesions in juveniles. Adult Chinook Salmon trapped for broodstock often have infections of *Saprolegnia spp.* that can cause pre-spawn mortality if not treated chemically (e.g., with formalin).

F. Various Myxobolid species

These species are microscopic multicellular parasites, like *Henneguya* spp., can be visualized at low to moderate numbers in kidney smears and other organs of Chinook Salmon beginning in the spring of their first year and continuing through smolt release the following year. As with other common or "normal" parasites, the contribution of infection to physiological stress and reduced immunity during non-optimal conditions is unknown.

G. Hexamita spp.

This parasite is a flagellated protozoan that is present in the intestine of Spring Chinook Salmon at all life history stages. This parasite causes gut inflammation at times but has not caused mortality.

IV. Potential future pathogens at Warm Springs NFH

A. Aeromonas salmonicida (A. sal.)

This is the bacterial species that causes Furunculosis. The organism is also highly temperature dependent. Disease outbreaks typically do not occur when the water temperature is below 13 °C (55 °F), but outbreaks are very likely to occur at temperatures above 20 °C (68 °F) and somewhat likely to occur between those two temperatures (13 - 20 °C) depending on other conditions. *A. salmonicida* has been isolated from both returning adult Chinook Salmon and juveniles held on station. The projected climate outlook could favor increased disease from this pathogen.

B. Aeromonas hydrophila complex spp.

These are bacterial species that cause Motile Aeromonas Septicemia (MAS) with clinical outcomes that include tail/fin rot and epidermal ulcers. This bacteria has been identified in adult and juvenile disease outbreaks at Warm Springs NFH in the past. These species are largely opportunistic and associated with poor water quality, warm temperatures, and other stressors. As summer temperatures increase above 16 °C (60 °F), these pathogens could become problematic at Warm Springs NFH.

C. Flavobacterium branchiophilum

This is the bacterium that causes Bacterial Gill Disease, but it has not been a significant disease issue at Warm Springs NFH. However, as the duration of low-summer flows increase and the magnitude of summer flows decrease, this disease may become more prevalent in the future.

D. Flavobacterium psychrophilum

This is the bacterium that causes Bacterial Cold Water Disease (BCWD) in salmonid fishes. The name "cold water" is somewhat misleading because this species of bacteria has been cultured from infected fish in water as warm as 20 °C (68 °F).

E. Nucleospora salmonis

This is a microscopic microsporean parasite that has been identified from salmonids in adjacent watersheds of the Warm Springs River. We do not know whether increased temperatures will increase the prevalence of this parasite in the Deschutes River Basin. However, as water temperatures during the summer elevate above 16 °C (60 °F), the immune systems of the fish are less likely to combat the parasite successfully if it is present.

F. Ichthyobodo spp.

This species is a single celled flagellate that infects juvenile Chinook Salmon at Warm Springs NFH. Mortality will increase and remain elevated until formalin treatment is applied. At lower water temperatures ($<10 \, ^\circ$ C or $<50 \, ^\circ$ F), formalin treatment can be applied once every one to two weeks to keep parasite levels sufficiently low to prevent daily mortalities prior to release of the fish. When water temperatures exceed 14 $^\circ$ C (57 $^\circ$ F), formalin may need to be applied as often as once every four days to keep parasite levels manageable. With projected increases in future mean temperatures, this parasite is likely to have a greater impact on Chinook Salmon at Warm Springs NFH, either resulting in increased use of formalin, higher mortality, or both.

G. Tetracapsuloides bryosalmonae

This species is the myxosporean parasite that causes Proliferative Kidney Disease (PKD), and has produced significant disease and mortality of salmonid fishes in Europe, British Columbia, California, Idaho, and Montana, and on private aquaculture farms in western Washington. The life cycle of the parasite includes an invertebrate bryozoan intermediate host that releases infective spores into the environment. Whether the bryozoan host is currently present in the Warm Springs River is unknown. Clinical diseases of salmonid fishes are temperature-dependent with significant disease and mortality at warmer temperatures (>14 °C or >57 °F). Warming water temperatures may also expand the distribution of PKD cases, either through pathogen migration or habitat expansion of its bryozoan intermediate host, or both. Surviving fish develop resistance to future infections of PKD. No treatment exists currently. With projected increases in water temperatures, PKD could become a significant pathogen and management challenge at Warm Springs.