

Causes and Consequences of Vateritic Otoliths in Hatchery-reared Coho Salmon

Background:

Although Coho Salmon (*Oncorhynchus kisutch*) are of cultural, ecological, and economic importance throughout their range, anthropogenic activities have led to widespread declines in natural populations. To mitigate such declines while sustaining salmon fisheries, federal, provincial, and private organizations have undertaken large-scale hatchery-rearing of juvenile Coho. Unfortunately, once released into the wild, hatchery-reared Coho juveniles survive at generally low rates compared to wild-origin Coho juveniles, such that hatcheries have proven much less effective than initially hoped, both ecologically and economically.

Recently, differences have been observed between the sagittal otoliths of hatchery-reared and wild-origin Pacific salmon. Sagittal otoliths are essential sensory structures that enable all teleost fish to hear and maintain balance. Sagittal otoliths are normally composed of aragonite, a polymorph of calcium carbonate, but otoliths with inclusions of vaterite, an abnormal polymorph, also occur. Although these 'vateritic otoliths' have been shown to occur in less than 10% of wild-origin salmon, they are extremely common in hatchery facilities, affecting 60-80% of hatchery-reared salmon. Vaterite formation is irreversible once begun and results in otoliths which are larger, lighter, more brittle, and less regularly shaped than their aragonite counterparts. Because of these differences, vaterite deposition likely reduces otolith function and causes severe hearing loss in salmon; potentially leading to modified behaviour and associated impacts on individual survival and population recovery.

Despite long-standing evidence of the occurrence of vateritic otoliths, the causes and consequences of their formation and prevalence in hatchery-reared salmon are largely unknown. Thus, the **goals** of our project are to determine the causes of vateritic otolith development in hatchery-reared Strait of Georgia (SoG) Coho Salmon and investigate the consequences these vateritic otoliths have on Coho Salmon predator avoidance behaviours and marine survival.

Objectives and Methods:

Objective 1. Investigate the prevalence of vateritic otoliths in Coho Salmon across SoG hatchery facilities, rearing years, life-stages, and stocks

We will investigate the prevalence of vateritic otoliths by comparing the frequency and % composition of vateritic otoliths between wild-origin and hatchery-reared Coho Salmon across differing hatchery facilities, rearing years, life-stages, and stocks. Preliminary results from previous years' collections suggest that vateritic otolith prevalence differs across hatchery facilities and years but in all cases, SoG hatchery-reared Coho have a much higher prevalence of vateritic otoliths than wild-origin Coho Salmon. We will collaborate with several Vancouver Island (Puntledge, Goldstream, Big Qualicum, Quinsam) and Fraser Valley/Vancouver (Chilliwack, Inch Creek, Capilano) Coho Salmon hatcheries to co-ordinate the collection of otoliths. In Spring 2021, we will receive the otoliths of ~60 hatchery-reared (adipose fin-clipped and/or coded-wire tagged) Coho Salmon smolts from DFO smolt health screenings (so we do not euthanize any additional fish). In Fall 2021 and Fall 2022, we will travel to each of these hatcheries to collect the otoliths of ~50 hatchery-reared and ~50 wild-reared returning Coho Salmon adults during broodstock collections. We will analyze the otolith pairs for vaterite frequency and % vaterite composition. We will repeat otolith collections from those smolts being released and adults returning at each of these hatchery facilities over the next four years every Spring and Fall (2021-2024).

Deliverables:

Document and compare the prevalence and differing amounts of vateritic otolith deformities occurring at differing hatchery facilities and across differing rearing years, life stages, and stocks.

Objective 2. Determine which hatchery-rearing practices cause vateritic otoliths to form in Coho Salmon

We will investigate the causes of vateritic otoliths in hatchery-reared Coho Salmon by determining which hatchery-rearing practices influence the frequency and % vateritic composition of otoliths. We will investigate these

causes in two different ways: i) through on-site rearing assessments across different SoG hatcheries, and ii) direct hatchery-rearing trials.

For the on-site rearing assessments, we plan to create a hatchery-rearing practice/conditions assessment audit form. This audit form will help us to distinguish the differences in rearing practices and conditions across differing SoG Hatchery facilities that Coho Salmon juveniles are exposed to. Differences may include tank design and rearing conditions, feed composition and feeding practices, handling techniques, and water quality parameters. We will collect data on the rearing conditions and practices employed at each facility and obtain facility-specific fish culture notes and data from each Salmonid Enhancement facility through collaboration with the hatchery managers. Once we have collected this information, we will be able to link specific hatchery-rearing practice/conditions to the frequency and % vateritic otolith composition seen in the otoliths collected from the Coho Salmon smolts from each facility (Objective 1). We will complete on-site rearing assessment audits for four years, each Spring from 2021-2024.

We began some preliminary direct hatchery-rearing trials in May 2020 at Goldstream Hatchery when Goldstream Coho fry were first ponded. Our trials include an investigation into how tank water flow direction, feed composition, environmental enrichment (the addition of plants, natural substrate, and structures to hide under/in) and tank density effects vateritic otolith frequency, asymmetry, and % composition. Otoliths are collected from a subsample of 20 fish from each rearing group weekly and analyzed for vaterite frequency and % composition. We plan to continue these trials until the Coho Salmon juveniles smolt and are released into the Goldstream river each Spring. If these hatchery-rearing practices are shown to influence vateritic otolith development and % composition, we will repeat these trials over the next 2 years with two additional parental brood years (May 2021-June 2022 and May 2022-June 2023). However, if we find that these hatchery-rearing practices do not influence vaterite otolith development and % composition, we will investigate a new set of hatchery-rearing practices (e.g., growth rate, noise exposure, water quality) in subsequent years. If we can successfully link hatchery-rearing practices to vaterite otolith development, we will be able to create recommendations on rearing-practices to help avoid such abnormalities in the future.

Deliverables:

Investigate and identify which hatchery-rearing practices seem to be causing vateritic otolith formation in hatchery-reared Coho Salmon from data collected from the on-site assessments and direct hatchery-experiments.

Objective 3. Investigate whether vateritic otoliths influence the predator avoidance behaviour of Coho Salmon

We will conduct laboratory experiments using predator sound playbacks in a large, insulated tank to examine whether vateritic otoliths influence the predator-avoidance behaviours of Coho Salmon smolts. We will collect control recordings (recordings without predator sounds) and experimental predator recordings (recordings with Pacific harbour seal sounds, a top predator of Pacific salmon smolts) using compact audio recorders connected to hydrophones. Hatchery-reared and wild-origin Coho Salmon smolts will be placed in and acclimatized to the experimental tank prior to the beginning of trials. Swimming behaviour (e.g., total travel, directionality, bottom time) and vocal behaviour (e.g., calling rate, call amplitude, call frequency) of individual fish in the experimental tank will be measured in the absence of predator sound (control playbacks) as well as during and after predator sound exposure (experimental predator sound playbacks). The use of predator sound playbacks in these trials allows for isolation of Pacific harbour seal sounds as a stressor independent of visual cues and predator movement effects. Once the trials are completed, the fish will be euthanized, and the otoliths will be analyzed for vateritic otolith frequency and percent composition.

Deliverables:

Determine whether Coho Salmon smolts possessing one or two vateritic otoliths will exhibit lesser behavioural reactions to the experimental predator sound playbacks compared to the Coho Salmon smolts possessing two aragonitic otoliths.

Objective 4. Investigate whether vateritic otoliths influence the survival rates of Coho Salmon

We will investigate whether vateritic otoliths influence the survival rates of wild-origin and hatchery-reared Coho salmon juveniles and adults at Goldstream hatchery (Objective 2). We will pit-tag subsamples of the Coho Salmon smolts that we are rearing at Goldstream hatchery under differing rearing-conditions (tank flow direction, tank density, environmental enrichment, and feed composition) and release them in Spring. We will install a pit-tag array in Goldstream river estuary to capture all returning pit-tagged adults. We will compare the number and % composition of vateritic otoliths of Coho smolts released in Spring 2022 to the number and % composition of vateritic otoliths of Coho jacks/adults returning in Fall 2022/2023. If we can link the smolts to adult return rates with those specific experimental rearing-practices used at Goldstream hatchery, we may be able to link the proportion of vaterite and survival with hatchery-rearing practices. A relationship between the prevalence of vateritic otoliths and Coho Salmon returns will have implications for restoration efforts, aquaculture, and the recreational and commercial fishing industries.

Deliverables:

We will gain estimates of Coho Salmon marine mortality/survival correlated with vateritic otoliths and associated hatchery-rearing practices.

Significance:

The proposed research will be the first to investigate the causes of and impacts vateritic otoliths have on Pacific salmon stocks. This research will help managers to evaluate the effectiveness of current hatchery-rearing efforts and determine which hatchery-rearing conditions should be implemented to reduce the prevalence and formation of vateritic otoliths in Coho Salmon juveniles. Furthermore, determining whether vateritic otoliths impact the marine survival or predator-avoidance behaviours of Coho Salmon could increase the generally poor success rates of hatchery restoration efforts. This, in

turn, may lead to faster recovery of Pacific salmon stocks, paving the way for sustainable development of coastal industries including recreational and commercial fisheries, tourism, and international commerce. This research will contribute to the fields of fish welfare and fisheries science by adding to our knowledge of Pacific salmon behaviour and physiology.