

**The Hakai Institute Juvenile Salmon Program: Early Life History Drivers of Marine Survival in Sockeye, Pink and Chum Salmon in British Columbia, Canada**

by

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**Keywords:** early marine survival, feeding biology, growth, pathogens and parasites, migration dynamics

## **Abstract**

The Hakai Institute Juvenile Salmon program is an ongoing initiative that was established in 2015 in partnership with the University of British Columbia, University of Toronto, Simon Fraser University and Salmon Coast Field Station. This program researches the early life history of juvenile salmon in coastal British Columbia. Primary research objectives are determining: 1) Migration timing rates and routes; 2) Migration habitat, including physical and chemical oceanographic conditions, and availability of plankton prey; 3) The impacts of prey phenology, quantity and quality on juvenile salmon growth and condition; 4) Species and stock specific feeding biology and competitive interactions; 5) Pathogen and parasite infection dynamics; and 6) Mortality estimates. The program targets Fraser River sockeye, and pink and chum salmon, but additionally provides information on coho, chinook and herring through incidental capture. The field program operates between May and July during the peak of the juvenile sockeye outward migration. Purse seine and oceanographic sampling is conducted in the northern Strait of Georgia / Discovery Islands region (~ 220 km from the Fraser River mouth) and the Johnstone Strait / Queen Charlotte Strait region (~ 180 km from the northern Strait of Georgia). As such, this program informs early life history across two critical legs of the Fraser salmon northward migration.

## **Introduction**

Fluctuating adult returns and recruitment have been observed across numerous salmon species and populations in southern British Columbia (BC). Chinook, coho and steelhead have all returned in increasingly low numbers in recent decades (Riddell et al., 2013; Scott, 2008; Zimmerman et al., 2015), while pink salmon have returned in unprecedented high numbers (Irvine et al., 2014), and Fraser River sockeye have been in steady decline, with some notable exceptions, for almost two decades (DFO, 2015). Of the Pacific salmon, sockeye are arguably the most important culturally and economically. The lowest return of Fraser River sockeye was recorded in 2009 which prompted a Federal Commission of Inquiry into the decline of sockeye salmon, commissioned by Justice Bruce Cohen. The Cohen Commission issued 75 recommendations for government regarding Fraser River sockeye salmon of which several focussed on improving understanding of the early marine phase (Cohen 2012a, b). A prior evaluation of drivers of declines in sockeye salmon stocks in BC abundance identified the early marine phase as critical to salmon stock recruitment (Peterman et al., 2010).

Multiple factors are expected to impact juvenile salmon survival during the early marine phase, with food availability for growth considered to be the leading contender, followed by pathogen and parasite infection (Cohen, 2012b; Peterman et al., 2010). Strong correlative evidence supports the role of feeding conditions in salmon stock fluctuations (Beamish et al., 2012; Thomson et al., 2012), yet few studies have examined plankton prey phenology, quality, and quantity and explicitly linked this to the growth and condition of fish. An additional area of uncertainty and research priority is the role of pathogens and parasites in the survival of salmon during their early marine phase (Cohen, 2012a; Cohen, 2012b; Cohen, 2012c).

Fraser River sockeye experience a wide range of conditions along their juvenile outmigration route. The majority of juvenile salmon exit the Strait of Georgia through the Discovery Islands and Johnstone Strait into Queen Charlotte Sound (Figures 2–4). Oceanographic conditions differ considerably across these regions, affecting plankton productivity and hence food availability for the juvenile fish. The Strait of Georgia basin (Salish Sea) and Queen Charlotte Sound are typically stratified and exhibit high primary production during the spring and summer months (Jackson et al., 2015). Conversely, the Discovery Islands / Johnstone Strait region is subject to high tidal mixing which is expected to reduce primary production through light limitation, concomitantly supporting lower zooplankton biomass (McKinnell et al., 2014). Research conducted by the Hakai Institute from 2015 to 2016 has confirmed that this region does indeed support lower zooplankton biomass (Mahara et al., in prep). Growth hormone (IGF1) data have provided first evidence for poor juvenile salmon growth in this region (Ferriss et al., 2014), however, further research is required to establish the impacts of migration through the Discovery Islands / Johnstone Strait region on juvenile salmon growth, health, and ultimately survival.

Poor feeding environment in the Discovery Islands / Johnstone Strait make this region a potential choke point for out-migrating juvenile salmon that may determine their capacity to survive changes in productivity in the Strait of Georgia and / or Queen Charlotte Sound. The Strait of Georgia and / or Queen Charlotte Sound are critical to the early life history of juvenile Fraser River salmon (and other salmon stocks exiting into the Strait of Georgia) as they spend approximately their first 6–10 weeks at sea migrating through these regions (Preikshot et al., 2012; Welch et al., 2011). Productivity in these regions, and on the BC coast in general, is subject to variability associated with natural climate cycles (e.g., ENSO, PDO) and long-term climate change (Riche et al., 2014). Related changes in the timing of spring plankton blooms (Allen and Wolfe, 2013) and the quantity and quality of subsequent summer plankton growth (El-Sabaawi et al., 2009) are expected to be key factors in determining the prey fields available to juvenile salmon and their growth, condition, and survival.

The Discovery Islands / Johnstone Strait region hosts a large portion of British Columbia's salmon farms. Since the primary migration route for wild Fraser River salmon is through this region there is high potential for pathogens and parasites to be transmitted between wild and farmed fish (Cohen, 2012b; Cohen, 2012c). Parasites and pathogens can be a source of significant mortality in wild salmon (Miller et al., 2014) and can cause catastrophic losses in salmon aquaculture (Mennerat et al., 2010). However, our current understanding of infectious disease dynamics in wild and farmed fish, and their interactions, is limited. There are a large number of pathogens that naturally infect wild Pacific salmon (Miller et al., 2014), and the domesticated environments in aquaculture may change the population dynamics and phenotypic traits of pathogens in ways that may threaten both wild and farmed salmon (Mennerat et al., 2010). Thus, understanding the ecology and evolution of pathogens in this context is essential for the sustainable management of both wild and farmed salmon in British Columbia.

Pathogens and parasites may have both direct (lethal) and indirect (sub-lethal) effects. In addition to having the potential to cause direct mortality, pathogens can compromise the ability of salmon to grow (Godwin et al., 2017; Sandell et al., 2015), compete for food (Godwin et al.,

2015), and avoid predators (Krkošek et al., 2011; Mesa et al., 1998). Because these indirect effects can reduce foraging success (Godwin et al., 2017; Godwin et al., In press) they would be expected, if occurring, to amplify the poor feeding conditions in the Discovery Islands / Johnstone Strait. This highlights the importance of understanding infection dynamics in addition to feeding conditions in the region. Infection dynamics are expected to depend on exposure to pathogens outside of the region, as well as a combination of wild fish migration paths, residence time in the vicinity of farmed fish net pens, and factors influencing transmission, such as the persistence of pathogens and parasites in the water column (Miller et al., 2014).

Stock-specific run timing and migration routes add an additional layer of complexity to understanding salmon early marine survival. Run timing interacts with plankton bloom timing and can be critical to the feeding conditions experienced by juvenile salmon (Chittenden et al., 2010). Migration routes and rates determine the extent to which specific populations interact with feeding hotspots / barrens as well as potential sources of pathogen transmission (e.g., sympatric wild host fishes and / or aquaculture facilities). As such, knowledge of stock specific migration behavior is essential to inform stock specific risk assessment and facilitate the development of management strategies to, for example, reduce farm-wild fish interactions if needed.

## **Program objectives**

**Objective 1:** Evaluate the controls of prey phenology, quantity and quality for migrating juvenile salmon in the northern Strait of Georgia, Discovery Islands and Johnstone Strait. Specifically:

- Characterise the physical and chemical oceanographic environment through comprehensive high-frequency measurements;
- Measure the spatial and temporal dynamics of phytoplankton and zooplankton biomass, species composition and production;
- Determine the physical and chemical drivers of plankton composition, stoichiometry and lipid content which define their quality as food, and their small-scale spatial and temporal variability.

**Objective 2:** Determine the stock-specific migration behavior of juvenile sockeye salmon, and co-migrating salmon species, through the Discovery Islands and Johnstone Strait with respect to:

- Migration rate from the northern Strait of Georgia to Queen Charlotte Strait;
- Migration route through the Discovery Islands and Johnstone Strait;
- Migration timing.

**Objective 3:** Determine juvenile salmon feeding biology and measure growth and condition across a spatial-temporal gradient of prey quantity and quality. Specifically, determine:

- Spatial variation in juvenile salmon prey fields and the importance of migration routes in determining diet and foraging success;
- Interaction of plankton phenology and stock specific run timing in determining diet and foraging success;
- Trophic niche and competitive interactions among juvenile sockeye, chum, and pink salmon across a high to low production gradient;

- Role of prey quantity and quality in determining the early marine growth and condition of juvenile salmon.

**Objective 4:** Determine juvenile salmon parasite and pathogen infection dynamics across the Discovery Island / Johnstone Strait region with respect to:

- Spatial and temporal dynamics of stock-specific parasite and pathogen load through the Discovery Island / Johnstone Strait region;
- Spatial and temporal dynamics of stock-specific immune and stress related gene expression, and their relationships to condition and pathogen load;
- Disentangling the relative and cumulative contributions of feedings conditions and parasites / pathogens to juvenile mortality.

**Objective 5:** Estimate the mortality rates of juvenile salmon during their Strait of Georgia to Queen Charlotte Strait migration.

### **Program history: 2015–2018**

The Hakai Institute Juvenile Salmon Program was established in 2015 in partnership with the University of British Columbia, University of Toronto, Simon Fraser University and Salmon Coast Field Station. The field program operates between May and July during the peak of the juvenile sockeye salmon outward migration. Sampling occurs in two locations: 1) the northern Strait of Georgia / Discovery Islands – operated by the Hakai Institute Quadra Island Field Station and; 2) Johnstone Strait / Queen Charlotte Strait – operated by the Salmon Coast Field Station based on Gilford Island (Figures 2–4). In 2015 and 2016, juvenile salmon sampling had wide spatial coverage in both regions in order to closely examine small-scale regional variability (Figure 2 and 3). In 2017, the extent of juvenile salmon sampling was reduced to target key entry points into the Discovery Islands region from the Strait of Georgia and exit points from Johnstone Strait into Queen Charlotte Strait (Figure 4). This focused sampling strategy informs migration and health parameters for fish after passage through 1) the Strait of Georgia (~ 220 km) and; 2) the Discovery Islands / Johnstone Strait (~ 180 km). The focused sampling strategy adopted in 2017 will be implemented in 2018 and subsequent years of this research and observation program.

### **Field methodology**

Sampling sites were visited every 4–7 days in 2015 and 2016 and weekly in 2017 to collect fish using purse seine nets (bunt: 27 m × 9 m with 13 mm mesh; tow: 46 m × 9 m with 76 mm mesh; Figure 1) deployed from 6–8m twin-outboard motor vessels. The size of the sampling vessels allows ready access to a wide range of habitats, including those near to shore at depths ≥ 10 m. The purse seine effectively samples sockeye (*Oncorhynchus nerka*), pink (*O. gorbuscha*), and chum (*O. keta*) that school together, and incidentally samples coho (*O. kisutch*), chinook (*O. tshawytscha*), and Pacific herring (*Clupea pallasii*).

Each site is spatially defined using landmarks that demarcate a 1 nm reach of shoreline and extend perpendicularly to approximately 250 m offshore. Fish are sampled within this site and the exact location of capture is recorded for each seine. During each sampling event we first

conduct a visual survey transect of the site to enumerate surface activity of juvenile salmon. Observers group their observations into counts within schools of surface activity. A school is defined as a cluster of surface activity that is separated from other clusters by a gap of low or no surface activity that is at least as large as nearby clusters. Counts are rounded to the nearest 0, 1, 10, 100, or 1000 surface events. This is classified as ‘site surface activity’. If no surface activity is observed after 20 minutes the seine is not set at that particular location (Figure 1). If surface activity is observed during that survey, the boat operator identifies an appropriate school to set on based on surface activity and conditions. Set surface activity is observed before the net is set while the operator positions the boat, and is determined by consensus among all observers and is rounded to 0, 1, 10, 100, or 1000 surface events. Non-targeted sets rarely catch fish, likely due to the size of the net. However, visually targeting surface activity yields high sample success rates. The efficacy of this approach has been validated in prior studies (Godwin et al., 2015; Price et al., 2013).

After the seine has been set, the net is hauled onto the boat by hand until the bunt forms a pocket in the water beside the boat. Enough water is maintained around the captured fish to minimize contact with the net. Fish are individually scooped up using a 4 L plastic jug and transferred to a 532 mL Whirl-Pak® Write-On sample bag where they are euthanized using a  $250 \text{ mg} \cdot \text{L}^{-1}$  concentration of tricaine methanesulfonate (TMS, MS-222). The sea water and TMS solution are then drained from the sample bag by perforating the bag with a push pin and the fish then transferred to a dry-shipper with vapour nitrogen to flash freeze them at  $-196 \text{ }^{\circ}\text{C}$ . Up to 30 sockeye, 10 chum, 10 pink, 10 coho, 5 chinook, and 10 herring are retained from each seine. The remaining fish are released after holding for no more than two hours (or before two hours if signs of fish stress are apparent). Prior to release, the number of fish remaining in the bunt is estimated by averaging the estimates generated by two team members who count groups of 10 or 100 fish at a time until all the fish in the net are enumerated. In 2017, several calibrations were conducted in which abundance was first estimated, and then enumerated completely by releasing fish slow enough to count them one at a time to confirm the accuracy of this method. Species composition was counted for a subset of fish from all depths and extrapolated to the total catch.

In conjunction with the purse seine program, oceanographic data are collected through the study area. Water column properties are measured with an RBR Conductivity-Temperature-Depth (CTD) Profiler harnessed with a Wetlabs Fluorometer. Zooplankton net tows are performed using a 50 cm diameter 250  $\mu\text{m}$  mesh ring net to characterize the prey fields. Zooplankton samples are preserved in a buffered 4 % formaldehyde seawater solution. Bottle samples are taken from multiple depths to collect water samples for nutrient concentrations, chlorophyll-a biomass and particulate organic matter (POM). Nutrient samples are analysed using a Lachat Instruments QuikChem 8500 Three Channel Flow Injection Analysis System at the University of British Columbia. Chlorophyll-a samples are analysed using a Turner Trilogy fluorometer using the JGOFS protocol (Holm-Hansen and Riemann, 1978). POM samples are analyzed for Carbon and Nitrogen stable isotopes at the University of California-Davis. The Juvenile Salmon Program is also extensively supported by the Hakai Institute Oceanographic Program, operating out of the Quadra Island Field Station. This program makes year-round observations of ocean conditions in the northern Strait of Georgia and Discovery Islands.

In 2017, acoustic tagging was introduced to the Hakai Institute Juvenile Salmon Program to contribute to research into the migration routes and migration rates used by juvenile Fraser River sockeye salmon and to develop mortality estimates. This research component is supported by the acoustic telemetry arrays presently maintained by Ocean Tracking Network and Kintama Research Ltd (Figure 2–4). Acoustic tagging will continue in 2018.

### **Laboratory methodologies**

Fish are dissected at the Hakai Institute Quadra Island Field Station. The Hakai dissection protocol is based on the protocol developed at the Molecular Genetics Laboratory at Fisheries and Oceans Canada's Pacific Biological Station, supervised by Kristi Miller. Hakai technicians received training at the Pacific Biological Station in 2015 to develop a dissection protocol that would facilitate tissue analysis using the Fluidigm Biomark platform (Jeffries et al., 2014). Due to the variety of questions being addressed in the Hakai program, this laboratory processing and dissection protocol was expanded to include methods for enumerating parasites and collecting tissues for stable isotopes, fatty acid profiling, parasite enumeration, growth measurements, and feeding biology.

A summary of sample processing steps is provided in Figure 5. Samples are transferred from the liquid nitrogen dry shippers directly into  $-80^{\circ}\text{C}$  storage in the lab. Aseptic technique is used throughout the dissection protocol. All fish contact areas are sterilized first by wiping surfaces first with a 10 % by volume bleach solution that contains 6–9 % sodium hypochlorite, then a rinse and wipe-down with smart2pure™ to remove the bleach, and finally a wipe down with 95% non-denatured ethanol. Dissection tools are first brushed with 'MILTEX™ EZ-Zyme All Purpose Enzyme Cleaner', then rinsed with smart2pure™ water, and finally autoclaved. All common surfaces in the lab are wiped down each day using Virox™ viricide wipes. One set of sterilized dissection tools is used to collect external muscle, gill and brain tissues, and to make a ventral incision to access internal organs. A different set of sterile tools is used internally to collect spleen, liver, heart, stomach, and kidney samples to avoid cross-contaminating internal organs with microbes from external tissues.

During dissections, fish are brought directly out of  $-80^{\circ}\text{C}$  storage and samples that need to remain frozen are dissected first while the fish is still frozen. Muscle, gill, brain, spleen, liver heart, and kidney samples are placed directly into a Bcision™ CoolBox XT containing dry ice that keeps samples frozen at  $-78^{\circ}\text{C}$  before being transferred back to  $-80^{\circ}\text{C}$  freezers, thus maintaining a cold-chain. Additional muscle tissues are taken for stable isotope analysis (stored at  $-20^{\circ}\text{C}$ ), fatty acid profiling (stored at  $-80^{\circ}\text{C}$ ), and RNA:DNA measurement of growth rates (stored at  $-80^{\circ}\text{C}$ ). Otoliths are extracted using plastic forceps, receive a smart2pure™ water rinse, and subsequently dried and stored separately in polyethylene tubes. Stomach samples are extracted whole by making an incision in the oesophagus and near the vent, extracting the stomach, and removing the gall bladder and extra liver tissue; they are then placed into a Falcon™ tube with 45 mL of 95 % non-denatured ethanol. Five scales are collected from above the lateral line and are adhered to scale-book paper and stored dry. Motile sea lice (i.e., pre-adult and adult) are removed from the fish and identified to species and stage under a dissecting microscope. We categorize and identify lice as *Caligus clemensi* pre-adult females, females, gravid females and males, and *Lepeoptheirus salmonis* pre-adult females, females, gravid

females, pre-adult males, and males. All fish are weighed to the nearest 0.1 g and measured for fork length and standard length to the nearest millimeter.

## **Program Reporting**

In an effort to provide useful information to the salmon research community, in-season reports are produced during the sockeye migration period. Reports include key monitoring components such as migration timing, species abundance, species composition, lengths, weights, parasite loads, and relevant oceanographic parameters. Program data are available on request in the form of well-documented and citable data packages. To request access to program data and to obtain links to in-season reports, visit <http://dx.doi.org/10.21966/1.566666>. Annual time series reports will be submitted to future NPAFC documents to rapidly share program results in a citable report.

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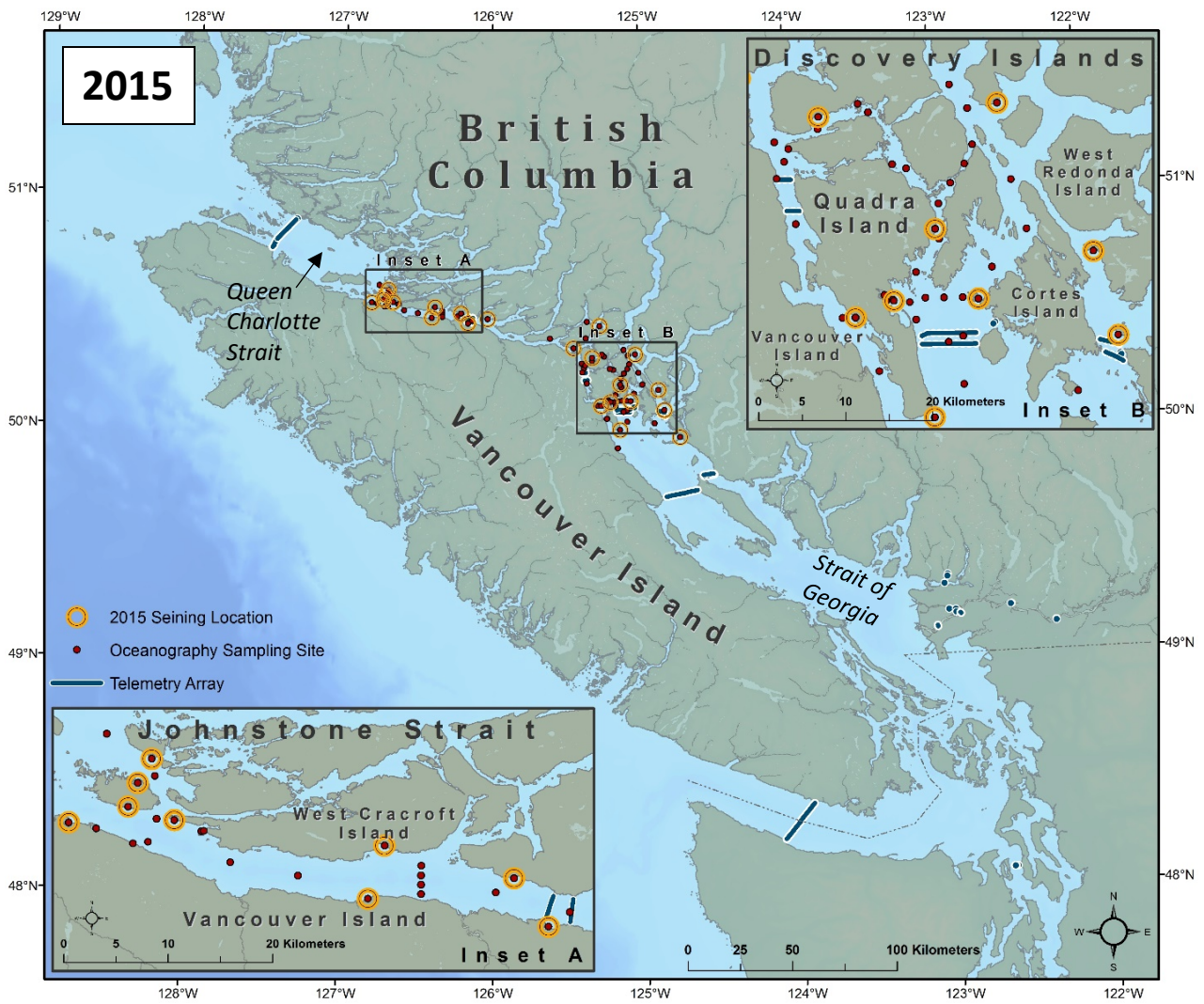
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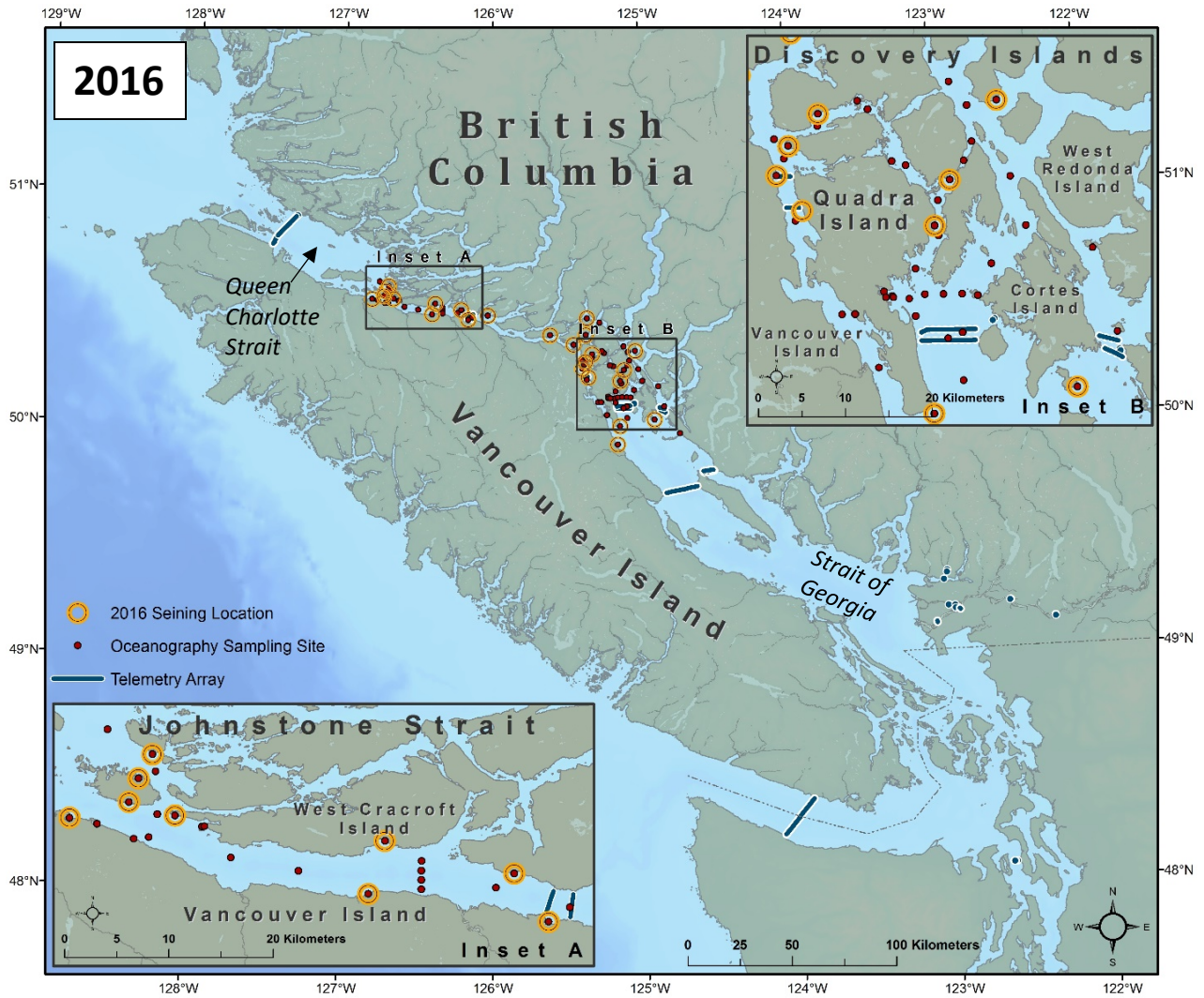
## Figures 1–5



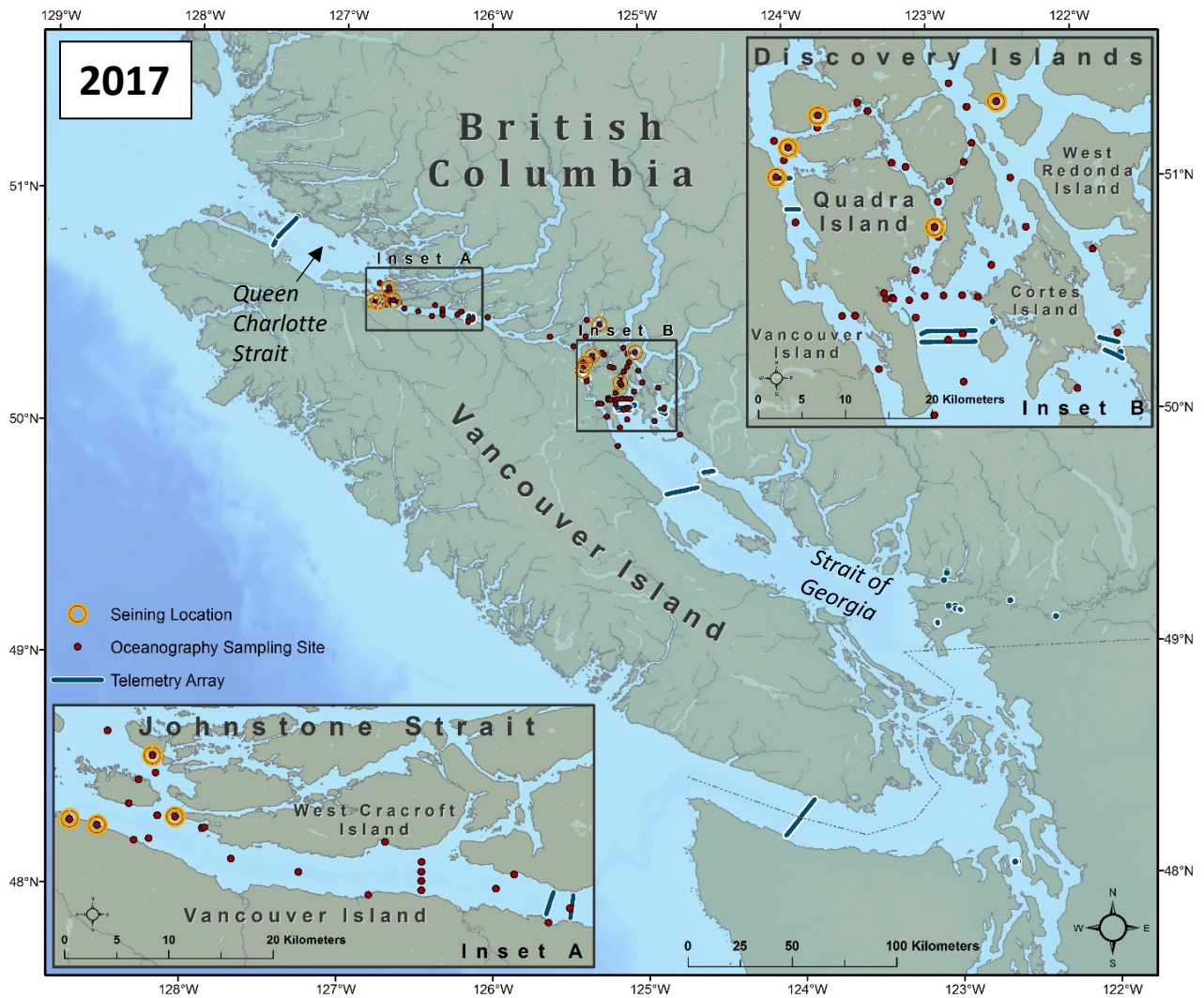
**Figure 1.** Deployment of purse seine net (bunt: 27 m × 9 m with 13 mm mesh; tow: 46 m × 9 m with 76 mm mesh) during the Hakai Institute Juvenile Salmon Program.



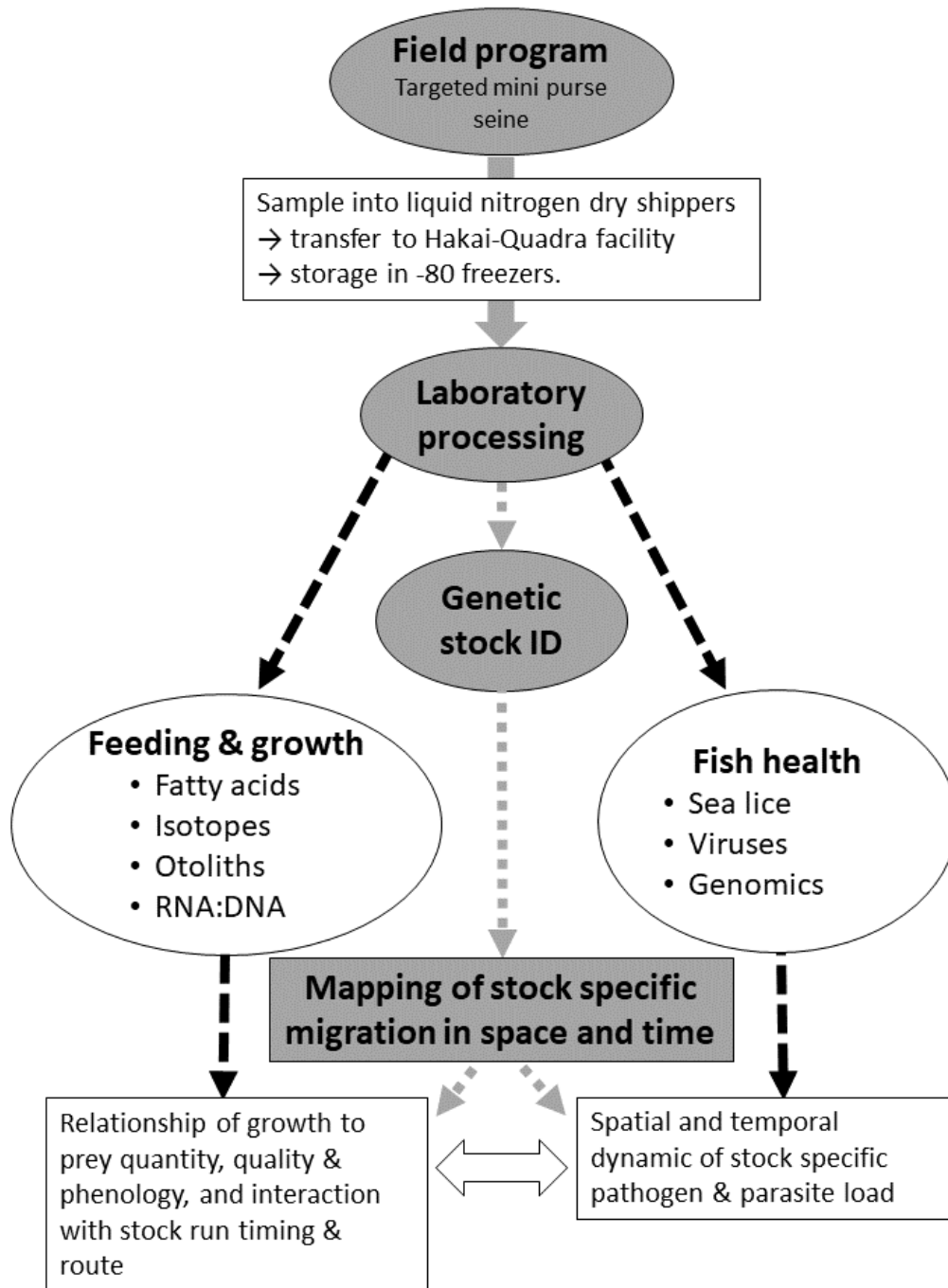
**Figure 2.** Map indicating the location of purse seine sites, oceanographic sampling sites, and acoustic telemetry arrays in 2015.



**Figure 3.** Map indicating the location of purse seine sites, oceanographic sampling sites, and acoustic telemetry arrays in 2016.



**Figure 4.** Map indicating the location of purse seine sites, oceanographic sampling sites, and acoustic telemetry arrays in 2017. The same sites are being used in the 2018 field program.



**Figure 5.** Schematic illustrating an overview of the samples collected from each salmon specimen during the Hakai Juvenile Salmon Program laboratory processing.