Department of Fisheries and Oceans Salmon Enhancement Program

Nitinat River Salmon Hatchery

Fish Health Management Plan

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1.1 Objective

In 2005, the National Aquatic Animal Health Program (NAAHP) was implemented. The purpose of the NAAHP is to reduce incidence and transmission of infectious agents at all levels of fish culture to meet international aquatic animal health management standards that are required to protect Canadian aquatic resources (wild and farmed).

With the NAAHP requirements in place, this Fish Health Management Plan provides best management practice guidelines for maintaining optimal health conditions for cultured fish to meet the current NAAHP requirements. All Salmonid Enhancement Program (SEP) facilities and those facilities partially funded or associated with DFO must maintain an up-to-date Fish Health Management Plan (FHMP) specific to their facility

This document should be reviewed annually by staff to ensure that it is current and changes should be made as necessary.

1.2 Target Audience

This document is intended for use by the fish culture staff at each SEP (or DFO associated facility) site for managing fish health and enabling an informed fish health decision making process. This document also serves as a valuable staff training tool.

1.3 Document Structure

Sections 1 and 2 contain general statements applicable to the operation of all Fisheries and Oceans Canada hatcheries involved in the enhancement of Pacific salmon in British Columbia.

Section 2 outlines the general principles of fish health management:

- Keeping the fish healthy and maintaining an optimal environment
- Keeping pathogens out
- Keeping disease from spreading
- Maintaining good records of appropriate information
- Minimizing impacts on natural populations
- Minimizing impacts on the receiving environment

Section 3 provides a brief overview of this particular facility.

Section 4 details the Standard Operating Procedures (SOPs) for fish health management practices for Pacific salmon culture that are specific to this facility.

Note: The focus of our work is the production of juvenile Pacific salmon for stock enhancement purposes. Netpen holding is limited to a handful of our facilities, which have the infrastructure and historical evidence of improved survival following a brief period of acclimation to a semi-natural environment. Additionally, this production strategy allows imprinting to a watershed for the eventual return in support of recreational fisheries in the areas whose natural spawning and rearing habitats are compromised.

Specific netpen practices should be included in the Section 4 SOPs where appropriate. Where indicated, Appendixes are included containing ancillary documents pertinent to the operation of the specific facility.

A glossary is provided in the Appendices to assist in defining terms that may not be familiar to staff.

Revisions to this document should be recorded to ensure the ability to track modifications to procedures.

1.4 Fish Health Management Team: Personnel duties and responsibilities

The Fish Health Management Team is comprised of the entities as defined below. The authority to alter the Standard Operating Procedures contained within this document lies with the Fish Health Management Team and should occur in a consultative process. The responsibility for carrying out the procedures defined within this document correctly and according to the individual protocol lies with the staff who have been trained in the individual procedures.

1.4.1 *Hatchery Management*

The hatchery managers are responsible for identifying and managing disease-related risk factors to minimize their impacts on fish health. The hatchery managers consult with the Veterinarian and DFO biologists on the management of fish health issues and is responsible for reporting outbreaks of significant diseases to other sites in the geographic vicinity and to the proper authorities.

1.4.2 *Hatchery Staff*

On-site staff is responsible for day-to-day fish health management, according to this Plan and the hatchery manager's directions.

1.4.3 *Support Biologists/Community Advisors*

Fisheries and Oceans biological support staff is available for consultation and to serve as a liaison between facility staff and the Enhancement Support and Assessment Unit.

1.4.4 *Veterinarian*

A licensed Veterinarian, in conjunction with facility and biological support staff, oversees fish health management for the SEP facilities. The Veterinarian, supported by the Pacific Biological Station Fish Pathology Laboratory, is expected to exercise good professional judgment in fish health matters. Specific duties include site visits, diagnostic workups for fish, treatment advice, and disease prevention and control recommendations. Where applicable, the Veterinarian will report disease findings to relevant authorities.

1.4.5 *Contact names and numbers*

Contact names and numbers for all key fish health personnel, including emergency numbers, are posted in an easily identifiable location at each site.

Contacts for this site:

Hatchery phone	250-745-3321
Hatchery fax	250-745-3416

Hatchery staff list:

Rob Brouwer, *Manager*	250-749-6760
Hans Galesloot, *Assistant Manager*	250-723-6403
Gary Dagley, *Enhancement Technician*	250-724-2790
Harley Gaetz, *Enhancement Technician*	250-723-2260
Ian Trepanier, *Enhancement Technician*	250-724-4491
Darrell Marshall, *Maintenance*	250-724-4946
Bill Cook, *Assistant Maintenance*	250-723-3858
Silvia Dean, *Administrative Assistance*	250-745-6261
Roy Bell	250-745-6261
Ed Filipchuk	250-724-5339
Gary Rooke	250-723-0652
Cory Jones	250-724-1930

Emergency Police, Fire Department and Ambulance	911
Non Emergency Police	123-456-7891
Non Emergency Fire	123-456-7891
Ambulance Service	123-456-7891

DFO CONTACT PHONE NUMBERS

DFO Community Advisor (Barry Cordocedo)	250-754-0329
DFO Community Advisor (Tom Rutherford)	250-746-5137
DFO Veterinarian (Dr. Christine MacWilliams)	250-729-8377
Fisheries Officer (Jim Robson - cell)	250-720-9906
Fisheries Officer (Rob Tompkins - cell)	250-720-9912
Observe, Record & Report Fishing/Habitat Violations	800-465-4336
Conservation Officer Service (Provincial Violations)	800-663-9453
Pacific Biological Station	250-756-7057
	250-756-7069

1.5 Definitions

Adipose fin: the small fin on the back of a salmonid, located between the dorsal fin and the tail. Excision of this fin provides a visual means to differentiate between wild and hatchery-produced fish.

Anaesthesia: A state of unconsciousness produced by anaesthetic agents, characterized by absence of pain sensation and varying degrees of muscle relaxation. This state is suitable for painful procedures like surgery, but requires greater monitoring as over-anaesthesia may be life-threatening if fish are unable to adequately respire.

Aneurysm: Weakness or injury to the wall of a blood vessel causing dilatation or ballooning and, in severe cases, threatening the integrity of the circulatory system resulting in haemorrhage or stroke. A weakened point of an artery, vein or the heart.

Aquacalm™: Metomidate hydrochloride is a useful sedative for broodstock transport and handling. This product is not intended for use in fish intended for human consumption; no withdrawal period has been established.

Biosecurity: Biosecurity refers to an integrated strategy to assess and manage the risks that threaten animal health, human health, food safety, and the environment.

Broodstock: a male or female breeding animal

CFIA – Canadian Food Inspection Agency

Clove Oil: This is a Generally Regarded As Safe (GRAS) human food additive containing the anaesthetics eugenol and isoeugenol, but whose pharmacology and metabolites are not well understood in fish. Neither clove oil nor its active ingredients are licensed for use in fish in Canada and its use is not recommended

CO_2: Carbon dioxide is a common anaesthetic in harvesting operations. As it naturally occurs in all animals, CO_2 is safe for the operator, the consumer and the environment and is not subject to a withdrawal time. However, hyperactivity is common with this chemical and it is difficult to reach deeper anaesthetic planes suitable for invasive procedures.

Coded wire tag: a wire tag imprinted with a binary code

Euthanasia: The deliberate ending of the life of an animal in an easy or painless manner.

Fork Length: The distance from the anterior aspect of the snout (or upper lip) to the tip of the medial caudal fin ray

Gametes: Male or female reproductive cells - the sperm or the egg

Hyperox™ : Trade name of a disinfectant chemical containing per acetic acid, acetic acid, hydrogen peroxide and a surfactant.

Isolation: Isolation refers to the separation of animals which have a specific infectious illness from those which are healthy and the restriction of their movement to stop the spread of that illness.

ITC: Introduction and Transfers Committee – an intergovernmental committee that regulates the movement of live aquatic animals throughout British Columbia

Margin of Safety: A measure of safety of a drug representing the range between the effective dose and the dose producing toxic effects. A low margin of safety indicates an increased risk of adverse side effects.

Micropyle: The specialized channel on an egg which enables a single sperm to swim down through the egg's surface and fertilize it

Moribund: fish in a dying state

Mort: a dead fish

Otolith: the bones of the inner ear of a fish. Controlled temperature manipulation during incubation of eggs or alevins will result in a distinguishable banding pattern on the otoliths. This technique is valuable for return stock assessments.

Outbreak: an unexpected occurrence of mortality or disease

Ovadine™: Trade name of a disinfectant chemical containing a buffered10% polylvinylpyrrolidone iodine (PVPI) solution in water. It may be used to disinfect equipment or fish eggs. Use on fish eggs is currently being monitored by Health Canada.

Perivitelline fluid: The perivitelline space is the area roughly between the yolk/embryo and the egg membrane. The perivitelline fluid fills this space.

Post-orbital length: The length of the fish from the posterior aspect of the eye to the end of the caudal base. The caudal base is found by moving the caudal fin laterally against the fish's body; a crease will appear at the junction of the hypural bones and the fin rays.

ppm: parts per million, equivalent measurement to mg/L (milligrams per liter)

Prevalence: the total number of cases of a specific disease in existence in a given population at a certain time

Quarantine: Quarantine refers to the separation and restriction of movement of animals which, while not yet ill, have been exposed to an infectious agent and therefore may become infectious. Strict separation of infected or potentially infected fish from healthy populations, including their products or items they might have contaminated, their effluent, feed and handling equipment, must occur for a period no longer than the longest incubation period for the infectious agent(s) of concern. Staff access is strictly controlled and requires heightened biosecurity measures. Release from quarantine requires veterinary approval.

Ripeness: having arrived at such a stage of development as to be ready to spawn

Rostral: the nasal/snout region

Saprophyte: An organism that commonly feeds on dead organic material, usually by decomposing and absorbing it, and assisting in its decay. Saphrophytes, in certain circumstances, may attack living hosts (eg, those weakened by primary pathogens or stress) and become pathogens.

Sedation: Chemical suppression of the central nervous system to allay irritability or excitement. Sedation is appropriate for minimally distressing events like transport, vaccination, marking, etc. and as a pre-

treatment prior to an anaesthetic agent. A drug that acts as a sedative at low doses may induce unconsciousness at higher doses.

Standard Length: The distance from the anterior aspect of the snout (or upper lip) to the end of the caudal peduncle (the caudal base).

Suppliers: any person or company that brings products to a site or removes something from the site. E.g. feed suppliers, mortality picks ups.

TMS: Tricaine methane sulphonate, an ester of benzocaine, is used as a fish anaesthetic. TMS is also commonly known as MS-222. A withdrawal period is mandatory if fish are to be released or used for human consumption.

Total Length: The length from the anterior aspect of the snout (or upper lip) to the posterior tip of the longest caudal fin ray when the caudal fin is spread in a 'natural' position

Vertical transmission: spread of a pathogen from the parent to the offspring

*Virkon™***:** Trade name of a disinfectant chemical containing peroxygen compounds, a surfactant, organic acids and an inorganic buffer system.

Withdrawal Time: The time interval after cessation of treatment before the animal or any of its products can be used as human food. Withdrawal times are based on the time interval required for tissue levels of the substance to fall below critical levels as decreed by legislation.

2 General Principles of Fish Health Management

2.1 Keeping Fish Healthy

Keeping fish as healthy as possible is critical to keeping pathogens from coming on site, reducing incidence of disease attributable to those pathogens already present, and/or minimizing spread of pathogens within or between sites.

Fish must be routinely monitored for signs of health and disease and for this reason all staff should be familiar with normal fish appearance and behaviour. Observations which may indicate a problem with the population include (but are not limited to):

- Physical changes – skin darkening, scale loss, fungal or ulcerative external lesions, increased opercular movements (respiration), protruding eyes
- Behavioural changes - loss of normal swimming and schooling behaviour, flashing, failure to elude capture, diminished response to feeding, gasping at the surface, clustering near water inflows or near airstones

Fish should be kept at reasonable densities as determined by species, size, number, type of rearing unit and water quality/availability. Changes in behaviour and physical condition should be reported to site management as early detection is the key to good disease management.

2.1.1 ***Maintaining an Optimum Environment***

2.1.1.1 Suitable Rearing Environment

The fish health staff is responsible for ensuring a suitable rearing environment for the fish at each life stage.

2.1.1.2 Monitoring Water Quality

Maintaining good water quality is vital to good fish health. The operator should maintain a regular program for monitoring and recording water quality at hatchery sites. Monitoring will vary between sites depending on location and the specifics of the aquatic environment and the frequency of monitoring will depend on available equipment and type of facility water use (i.e., flow through or recirculation). In-line monitoring may be applicable.

SOP: Water quality monitoring

2.1.1.3 Water Quality Contingency Planning

The facility should maintain a contingency plan in the event of acute deterioration of water quality (for example due to loss of flow or contamination of supply). Failure of pumps requires an immediate response. Systems should be suitably alarmed to indicate a water supply failure. The site should have backup systems to ensure water supply is not interrupted and quality is maintained.

SOP: Water quality contingency plan

2.1.2 ***Feed and Nutrition***

Feeding is both an art and a science. A site-specific, customized feeding program coupled with appropriately sized, high quality feed will fulfill the nutritional requirements needed for the growth and health maintenance of the fish. The amount fed will be influenced by many factors including: water temperature, species, body size, age, type of feed and different feed delivery methods.

Proper storage of feed is essential to maintain its nutritional value. Feed stored under improper conditions will result in rancidity and degradation of essential nutrients. Feed should be stored in secure buildings such that wildlife is excluded and spillage is prevented.

SOP: Feed, Feed Storage, & Feeding Practices

2.1.3 ***Common Fish Culture Procedures***

2.1.3.1 Anaesthetizing Fish

A number of fish health procedures require that fish be anaesthetized. Acquiring chemical anaesthetics requires a veterinary prescription. Netting of fish prior to anaesthesia should be done in as stress-free a manner as possible. Exposure to anaesthetic should be minimized while ensuring the anaesthetic level is adequate for the procedure. Anaesthetized fish should be carefully monitored at all times and the water quality of the anaesthetic bath – in particular, oxygen level – should be monitored.

SOP: Anaesthesia

2.1.3.2 Marking Fish

Marking fish is a valuable tool for accurate stock assessment. The species, number of fish to be marked and method of marking should be reviewed annually during this facility's production planning meetings. Marking should be done in a manner designed to result in minimal injury and stress to the fish. Appropriate anaesthesia and monitoring for adverse effects, both during the procedure and for several days following, are standard, as the stress of the procedure and resulting wound can compromise the immune response of the fish.

SOP: Marking Fish

2.1.3.3 Fish Transports

Fry, smolts and other life stages should be handled in as stress-free a manner as possible in preparation for transport. Equipment should be checked to prevent significant injury that could predispose fish to damage and/or disease. Proper hygiene and disinfection are adhered to. Appropriate transfer permits are obtained from DFO.

SOP: Transporting Fish

2.1.3.4 Vaccination

Vaccines are used to boost immunity to specific infectious diseases (e.g. Vibriosis) and are part of an integrated fish health management program. Vaccines are biological substances that must be stored (refrigerated) and handled as per manufacturer's instructions so as to maintain their safety and effectiveness. A product insert for each vaccine that is on site is kept in a safe, readily accessible place. Staff should be appropriately trained prior to undertaking the vaccination procedure to ensure that biologicals are used safely (i.e., wearing appropriate personal protective gear and taking suitable precautions).

Vaccination must be done in accordance with manufacturer's guidelines to ensure proper results. Since stress reduces the response of fish to a given vaccine, fish should be handled in as stress-free a manner as possible.

SOP: Vaccine handling, storage and administration

2.1.3.5 Euthanasia

In the uncommon situation where fish need to be euthanized, euthanasia should be done in as humane a manner as possible. The method used should result in rapid and irreversible loss of consciousness.

SOP: Euthanasia

2.1.3.6 Gamete Collection (Egg Take and Milt Collection)

At the Veterinarian's discretion, broodstock may be treated preventatively for specific infectious diseases prior to maturation to reduce the risk of vertical transmission of disease. Egg take and milt collection should be performed in as hygienic a manner as possible to prevent transmission of diseases to other broodstock and/or progeny. Adult fish should be anaesthetized and surface disinfected prior to gamete harvest and spawned adults should be euthanized as humanely as practicable. Carcasses are disposed of in a manner to prevent spread of disease. Males, if used multiple times, should be monitored for recovery from anaesthesia after each procedure.

SOP: Gamete Collection (Egg Take and Milt Collection)

2.1.3.7 Egg Disinfection

Eggs can be safely disinfected following fertilization and during water hardening.

SOP: Egg Disinfection

2.1.3.8 Egg Treatments

Developing eggs are sensitive to light and shock as well as fungal infections. Eggs are periodically checked for mortality, and presence of infectious diseases or fungus. Affected eggs should be treated as necessary.

SOP's: Egg Shocking, Picking & Egg Enumeration
Egg Disinfection

2.1.3.9 Juvenile release

The health and treatment status of fish is considered when planning intentional fish releases. The planned release of enhancement/conservation fish from our facilities will undergo a risk assessment to attempt to prevent undue harm to wild fish populations or public health. Fish are to be released in good health to minimize the transfer of pathogens to wild fish. The timing of release is also important to reduce stress and maximize survival of released fish.

SOP: Pre-Release or Transfer Disease Risk Assessment
Juvenile Release

2.1.3.10 Juvenile Treatments

There is a great deal of physiological stress associated with juvenile growth and smoltification. At the same time, the juvenile salmonid immune system is still developing. Because of this, juveniles represent a particularly susceptible life stage and judicious use of antimicrobial agents may help minimize losses due to infectious agents.

SOP: Juvenile Treatments

2.2 Keeping Pathogens Out

Biosecurity refers to an integrated strategy to assess and manage the risks that threaten animal health, human health, food safety, and the environment. The key components of a biosecurity program involve the exclusion of pathogens from a site and the containment of pathogens within a site if a disease situation does occur. The nature of enhancing wild populations using gametes collected from mature salmon returning from the oceans means that it is impossible to prevent the introduction of pathogens in all cases. Nevertheless, measures are in place to minimize the introduction of pathogens at key fish culture junctions and to minimize the impacts related to the presence of pathogens.

2.2.1 ***Site Physical Barriers***

Management is responsible for providing a suitable, secure rearing environment. Additionally, physical barriers to prevent uncontrolled or undesirable human and animal entry, the risks involved with movement of all personnel (staff, management, volunteers, Fish Health Management Team), visitors and equipment are assessed and managed.

SOP : Site and staff disinfection and biosecurity

2.2.2 ***Personnel Movement***

Staff will adhere to biosecurity procedures for the site. Where possible, personnel will not travel between hatcheries. If such travel is unavoidable, personnel will not return to a clean facility after visiting a

disease-suspect one, or will adhere to all biosecurity procedures at each facility to minimize the risk of inadvertently spreading disease between sites.

SOP : Site and staff disinfection and biosecurity

2.2.3 *Visitors*

Each site shall have posted procedures for all visitors, and visitors are expected to follow these procedures. Visitor access will exclude any areas containing sensitive life stages, i.e. incubation rooms.

SOP : Site and staff disinfection and biosecurity

2.2.4 *Predator Exclusion*

Every attempt should be made to exclude predators from the site. Predators should be excluded from the site. Predators include birds, rodents and occasionally mammals such as mink, river otters and bears.

SOP: Predator exclusion

2.2.5 *Suppliers*

Suppliers should be advised of operator and site procedures in advance. Suppliers who visit multiple sites shall be subject to strict biosecurity measures and may be requested not to come on site.

SOP : Site and staff disinfection and biosecurity

2.2.6 *Equipment Movement*

Where possible, equipment will not be shared between sites. This includes pumps, vehicles and fish handling equipment. Where this is not possible, equipment that must be used at multiple sites should be subject to strict biosecurity and disinfection measures between uses as per *2.2.7*.

2.2.7 *Equipment Maintenance*

To reduce the possible spread of pathogens by fish, personnel or via a waterborne route, equipment should be kept clean at all times. Equipment should be properly disinfected after each use and put away in its proper location.

SOP: Equipment disinfection

2.2.8 *Moving Fish Between Sites*

Fish movement between sites is kept to a minimum. A disease risk assessment should be performed in conjunction with the Fish Health Management Team prior to moving fish and necessary transfer permits should be obtained. Clinically ill fish will not be moved between sites. The move should be planned in advance to be as stress-free and short as possible. Fish should be transported as per *2.1.3.3* Particular care should be paid to the fish during transportation to avoid undue stress or possibility of escape. Water quality should be maintained and frequently monitored during transport.

The receiving sites will make arrangements for isolating the newly arriving fish. Once on site, measures should be used to limit the potential transmission of any previously undetected pathogens to the facility's original population.

SOP's: *Pre-Release or Transfer Disease Risk Assessment*
Egg and Milt Transport
Quarantine/Isolation Procedures for Suspected Disease Outbreaks

2.2.9 ***Broodstock Management***

The Veterinarian and/or Fish Health Management Team will develop specific disease screening procedures to minimize the risk of vertical transmission of pathogens from broodstock to eggs. Samples for disease screening must be collected in a sterile manner to minimize risk of contamination which can result in improper diagnosis.

Location of progeny from sampled fish should be tracked until such time as screening results have been received and reviewed by the Veterinarian and/or Fish Health Management.

For DFO enhanced fish, determining the causes of fish mortality prior to spawning can provide important information on disease incidence in the population and indicate the presence of vertically transmitted diseases.

SOP's:
Broodstock Selection
Broodstock Handling
Broodstock Treatments
Adult Carcass Disposal

2.3 **Keeping disease from spreading**

2.3.1 ***Separation of Fish Groups***

Different species or stocks are kept separated while on site. Rearing units are kept separate to prevent transmission of disease between groups.

2.3.2 ***Minimizing Disease Within the Site***

All efforts should be made to minimize disease on a site. All personnel will adhere to the facility hygiene and disinfection procedures as per *2.2.2*. Tank cleaning and moribund/mortality collection is carried out on a routine and frequent basis. This serves to reduce the potential exposure to pathogens and minimize predator attraction.

2.3.3 ***Monitoring Fish Health***

Fish should be monitored at least once daily for any unusual behaviour, visible lesions or other sign of disease. Changes in behaviour and physical condition should be reported to site management. Additionally, routine scheduled length/weight sampling during rearing allows a more detailed examination of the fish, as well as comparisons of actual versus expected gains and tracking of biomass per tank for appropriate density management.

SOP's: Juveniles-Health Observations
Individual Length/Weight and Bulk Weight Sampling Protocols

2.3.3.1 Mortality Classification

Mortalities should be examined for external signs of disease, as per the operator procedure, suspect mortalities may be examined internally. Suspected causes of mortality must be recorded and fish health management should be notified of any unusual numbers or types of mortalities.

SOP: Mortality Classification

2.3.3.2 Mortality Collection and Disposal

Mortalities should be collected on a routine and frequent basis to minimize the potential spread of disease, to minimize attractiveness to predators and to allow rapid identification of a health issue. The mortality storage area should be an appropriate distance away from any rearing units and outside usual travel corridors to minimize inadvertent spread of disease. Proper disinfection procedures should be adhered to after each mortality collection.

SOP: Mortality Collection and Disposal

The goal of good fish health management is to have healthy and productive fish. However if fish do become sick, they may require treatment with a therapeutant.

The Veterinarian maintains a Veterinarian-client-patient relationship with the operator that is the basis for disease diagnoses and prescribing treatments.

2.3.3.3 Medicated Feed: Handling, Storage and Inventory

Medicated feed should be stored in clearly marked bags separately from non-medicated feed. The storage area should be clean, dry and free of predators. The label on the medicated feedbag provides details about the feed, medication included, feed rate, name of the Veterinarian, prescription number and date it was milled.

Medicated feed should be inventoried separately from regular feed. Daily inventory records should be kept as the feed is fed to the fish according to prescription.

In the unlikely event that there is excess medicated feed after completion of the treatment, the Veterinarian should be contacted to determine proper handling and disposal.

SOP: Medicated Feed: Storage, Handling, and Feeding

2.3.3.4 Handling and Administering Medicated Feed

Medication mixed into feed has a Material Data Safety Sheet (MSDS) which specifies handling and safety precautions. An MSDS for all medications used on site must be on site in a readily accessible binder. All staff at this facility has undergone Workplace Hazardous Materials Information System (WHMIS) training and all chemicals must be handled safely; i.e., wearing appropriate personal protective equipment and taking suitable precautions for handling and disposal.

Medicated feed must be administered in accordance with the Veterinarian's instructions. The appropriate rearing unit(s) must receive the prescribed amount of medicated feed for the duration of treatment.

The Veterinarian must be informed if there is a lack of expected response within 5 days of the initiation of treatment.

SOP: Top-Coating Medicated Feed

2.3.3.5 Treatment Records

Provincial regulations require that treatment records for therapeutants include:

- Location of fish culture facility
- Species and stock identification
- Name of the prescribing Veterinarian
- A log naming the drugs (therapeutants), including
  - How they were administered
  - Treatment schedule including the date treatment commenced
  - Date of last treatment
  - Name and signature of the person responsible for administering each treatment

Detailed records of medicated feed administration are kept for the duration of treatment. Staff is responsible for monitoring for any adverse response to treatment (i.e., lack of appetite, lack of anticipated decline in morbidity and/or mortality levels) and reporting this information to the hatchery manager and the prescribing Veterinarian. Medicated feed records should be entered into ENPRO and a hard copy should be kept on site until the fish are released. In combination with inventory records, the fish receiving medication are readily identifiable during treatment and until the completion of the prescribed withdrawal time.

A copy of the treatment records will accompany those fish to another site if the fish are moved.

2.3.4 ***Fish Health Emergencies***

A fish health emergency is any situation where the health of the fish population is suddenly at risk. This may be due to a sudden, severe decrease in water quality or availability, or due to significant pathogens such as the IHN virus. Vigilant monitoring and early detection are the cornerstones of fish health emergency management.

2.3.4.1 System Failure/ Water Quality Event

If there is a system failure, all efforts should be directed to restoring sufficient water quality for the fish. Sufficient oxygen levels must be restored to support the fish. The site will immediately activate the Operator's Water Quality Contingency Plan, as per *2.1.1.3*. In the event of life-threatening poor water quality events, the fish should be taken off feed in order to decrease the oxygen demand and stress.

If an infectious disease problem is suspected, the operator Veterinarian and/or Fish Health Management must be ***immediately*** notified. If the problem is not easily discerned, event management and diagnosis will need to be done hand-in-hand.

2.3.4.2 Infectious Disease Emergencies

An outbreak is defined as an unexpected occurrence of mortality or disease. Not all outbreaks are fish health emergencies. Pathogens differ in many respects including ease of transmission, time until clinical signs of disease are apparent, severity of disease, and range of treatment options.

Accurate husbandry records and diligent monitoring of fish population health are central to the early identification of a disease situation. Rapid response is essential but should be determined on a case-by-case basis in conjunction with the Veterinarian and/or Fish Health Management.

Once an emergency has been recognized, certain steps are followed. The objective is to keep the pathogen "load" as low as possible and to prevent spread of the pathogen both within and off the site.

2.3.4.3 Emergency Response Steps

2.3.4.3.1 Quarantine

Quarantine is the enforced physical separation of the healthy population from a (potentially) infected population, their products or items they may have contaminated. At the Veterinarian's recommendation the site may be officially quarantined. Quarantine remains in effect until such time as the problem has been diagnosed and/or managed.

SOP: Quarantine/Isolation Procedures for Suspected Disease Outbreaks

2.3.4.3.2 Stop Fish Movement and/or Handling

The movement of all fish on/off and within the site may cease and fish will not be handled further. No visitors or non-essential staff is allowed on site unless previously authorized by Management.

2.3.4.3.3 Disinfection and Hygiene

Hygiene and disinfection on site, including procedures for personnel and equipment are strictly enforced.

SOP: Outbreak – Disinfection Protocols

2.3.4.3.4 Suppliers

In the case of an outbreak, suppliers (e.g., feed or oxygen delivery) are to be instructed to visit the site last or to make special arrangements.

2.3.4.3.5 Mortality Collection

The frequency of mortality collection is to be increased during an outbreak. Affected tanks are mort picked last and staff adheres to disinfection procedures between tanks and rearing units. If possible, separate gear is designated for the affected unit. All equipment, surfaces and clothing that come in contact with infected fish or potentially infectious material are thoroughly disinfected after use. Mortality collection and disposal procedures, as per 2.3.3.2, are strictly adhered to and provisions made for increased mortality pick-ups and disposal.

2.3.4.4 Determining the Cause of the Outbreak (Outbreak Investigation)

The Veterinarian may require records and appropriate sampling to determine the cause of the outbreak and best course of action. The Veterinarian and/or Fish Health Management will provide instructions for proper sampling. Water and feed samples may be requested. Samples must be properly handled, properly stored and promptly shipped as per the Veterinarian's or Fish Health Management's instructions to ensure prompt and effective analysis

Continued monitoring is required after the initial workup to determine the course of the outbreak and to assess whether treatment and/or management measures are effective. Frequent observations of fish are essential. Feeding response and water quality is monitored. All treatments and management changes are noted as they occur. The Veterinarian, Fish Health Management and site management will work together to review fish health records and make further management decisions. Any repeat sampling, including results, are duly noted.

SOP's: Outbreak Response

Sample Shipment to a Diagnostic Laboratory
Diagnostic Sampling protocols

2.3.4.4.1 Site Depopulation

Site depopulation is the total destruction of all animals on site in the event of a catastrophic outbreak. If site depopulation has been agreed upon, the procedure should be conducted as humanely as possible and in a manner consistent with principles of hygiene and biosecurity.

2.3.4.4.2 Reporting to Authorities

Where appropriate and/or in accordance with existing regulations, operator management will report the outbreak to Provincial or Federal authorities.

2.3.4.4.3 Communicating With Other Operators

The site management office will notify other operators in the geographic area of the outbreak.

2.3.5 ***Handling Drugs and Chemicals Properly***

2.3.5.1 Disinfectants

Disinfectants are stored in clearly marked containers. An MSDS for each disinfectant present on site is kept in a safe, readily accessible place, e.g., binder in the site office. As per WHMIS, all chemicals must be handled safely by trained staff e.g., wearing appropriate protective gear and taking suitable precautions.

SOP: Chemicals & Disinfectants: Supplies and Storage

2.3.5.2 Chemicals

Chemicals include, but are not limited to, fixatives such as formalin or Davidson's solution used for preserving fish tissues. These chemicals are stored in clearly marked containers. An MSDS for each chemical that is on site is kept in a safe, readily accessible place, e.g. binder in the site office. As per WHMIS, all chemicals must be handled safely trained staff e.g., by wearing appropriate protective gear and taking suitable precautions.

SOP: Chemicals & Disinfectants: Supplies and Storage

2.3.5.3 Biologicals

Biologicals are substances derived from animals or microorganisms that are used in the treatment, prevention or diagnosis of disease. Biologicals include vaccines, bacterins and antibody-based diagnostic tests. Enhancement hatcheries may use vaccines to boast the immune response to commonly encountered pathogens. Where applicable, these products are kept refrigerated and handled as per manufacturer's instructions. A product insert for each on-site vaccine is kept in a safe, readily accessible place. Trained staff must handle all biologicals safely e.g., by wearing appropriate protective gear as dictated by the MSDS and taking suitable precautions.

2.4 Keeping Good Records

2.4.1 ***Fish Health Records***

Fish health records include, but are not limited to:

- Inventory records

    - Includes source, number, location and lot of fish at the site
- Fish movement records
- Mortality records including clinical signs and mortality cause if known
- Diagnostic sampling records
- Diagnostic results
- Water quality records
- Therapeutics and medicated feed records
- Records of actions (other than therapeutics) taken to prevent or mitigate disease, e.g. refused shipment of potentially infected eggs
- Records of reporting to Provincial or Federal authorities, in accordance with existing regulation

Many of these records are computerized and form part of the integrated operator record keeping system. The operator will provide adequate system training and documentation to authorized site personnel including data entry and reports, e.g. ENPRO for DFO. Backups should be maintained.

Paper records not entered into a computerized system should be well organized, easily accessible and protected from damage, e.g. kept in binders.

Records should be kept for the duration of time the fish are on site. The operator will keep archived records at a suitable location in head office or securely stored off site. Records should be available for inspection upon request by BC MAFF.

Records should be reviewed on a routine basis by the operator Veterinarian and/or Fish Health Management Team to look for patterns in fish health and disease.

2.4.2 *Reporting to BC Fish Health Database*

The operator reports required fish health data, e.g. mortality cause and fish health event information to the BCSFA Fish Health Database on a monthly basis. Aquaculture companies keep records of data submission for audit by BC MAFF. Reporting to the BC Fish Health Database is also required of enhancement hatcheries and this data is also subject to audit by the BC MAFF. There is a shared responsibility to report what is occurring in fish culture regardless of the nature or purpose of culture. Wild and cultured fish share similar resources and compliance with the reporting requirements ensures that the maximum information is available to lead to informed and appropriate aquatic environmental and health management decisions.

2.4.3 *Egg Take Records*

Records should be kept for egg takes and broodstock disease screening. Records must accompany each shipment of eggs from the Broodstock location to the hatchery receiving the eggs, whether destined for onsite or off site incubation

2.5 Impacts on Non-Enhanced Stocks

2.5.1 *Fish Escape*

The Salmonid Enhancement Program intentionally releases cultured fish. Escapes in this context are less of a concern than for commercial producers using non-native or selectively bred stocks. However, infrastructure is in place to ensure fish escapes are discouraged. In the unlikely event that fish escape into nearby streams or watersheds, fish health records, including relevant diagnoses and treatments, must be made available to the appropriate regulatory authorities as required.

3 A Brief Overview of this Facility

Nitinat hatchery annually enhances Chinook, Chum, Coho and Steelhead stocks. Chinook stocks originate from the Nitinat and Sarita Watersheds. Chum stocks are primarily of Nitinat origin but recently have expanded to include fish from Grappler inlet near Bamfield. Coho and Steelhead are of Nitinat origin only.

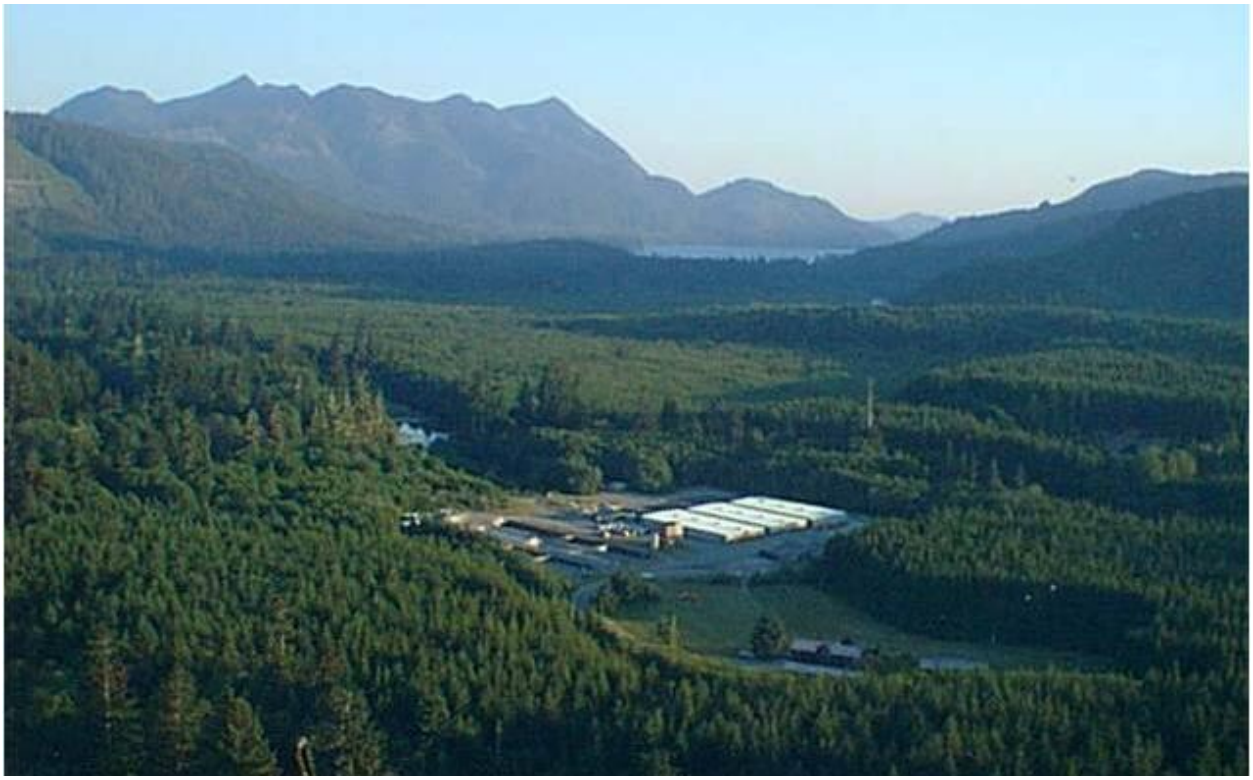
Stock & Species	Historical
Nitinat Chinook	6.0 million
Sarita Chinook	0.5 million
Nitinat Chum	35 million fed fry
Coho Smolts	250 K
Coho Fry	100 K
Steelhead Targets	15 K

Nitinat River Hatchery is situated near Nitinat Lake, a tidal salt water fjord, 70 kilometers southeast of Port Alberni on the west coast of Vancouver Island. Nitinat Lake is 23 kilometers long, 1.2 kilometers wide and has a maximum depth of 200 meters. A narrow tidal passage 3 kilometers long and 2.5 meters deep at low tide joins the lake to the Pacific Ocean. The waters of Nitinat Lake below 20-30 meters in depth are permanently anoxic with high levels of hydrogen sulfide. The facility produces chum, Chinook and coho, steelhead salmon.

The Hatchery annually performs the largest chum salmon egg take in all of Canada with a total of over 40,000,000 eggs being taken. The facility is a major contributor to the commercial chum fishery. Over a quarter of a million coho are produced each year as well as approximately 20,000 steelhead. It is also a major producer of Chinook, the annual release of which has grown to over 3,500,000 smolts. This has resulted in a thriving sport fishery inside Nitinat Lake and surrounding fishing locations on the West Coast of Vancouver Island. In addition, the hatchery fish are caught by Canadian and American commercial fishermen.

They are also involved with the Sooke, Toquart, Esquimalt and Goldstream salmon enhancement societies in the transplantation of Chinook and chum eggs. It also provides eggs for local classroom projects. Students involved in Salmonids in the Classroom education supplement their training with hands-on experience at the Hatchery.

The Hatchery personnel study enhancement techniques with the goal of improving the quality of smolts released. In association with the Ditidaht First Nations, the Hatchery monitors a program of downstream trapping, habitat availability and net pen rearing in a salt water tidal lake. They are also involved in stream enumeration, classification and mapping of spawning gravels. As Nitinat River has significant natural spawning populations, escapement monitoring is very important to preserve genetic diversity. The Hatchery's coded wire tagging program provides information on the distribution and timing of stocks, which is useful information to Fisheries' Management.




NITINAT RIVER
HATCHERY SITE
(28 HECTARES)
LITTLE NITINAT
RIVER
FISHWAY
PUMP HOUSE
NITINAT RIVER


1
2
3
4
5
6
7
8
9
10
11
12
13
14
Legend:
1, 2 & 3 – Keeper Channels
4 – Incubation
5 – Shallow Concrete Ponds #1-8
6 – Deep Concrete Ponds #9-12
7 – Adult Handling
8 – Small Containers
9 – Asphalt Raceways
10 – Power House
11 – Aeration Tower
12 – Offices
13 – Shop / Dry Storage
14 – Fishway


1
2
3
4
5
6
7
8
9
10
11
Legend:
1 – Cap Troughs
2 – Pet Troughs
3 – Mega Troughs
4 – Circular Tubs
5 – Deep Concrete Ponds#9-12
6 – Shallow Concrete Ponds #1-8
7, 8 & 9 – Keeper Channels
10 – Incubation
11 – Adult Handling

4 Standard Operating Procedures for Nitinat River Hatchery

The following list of Standard Operating Procedures outlines fish culture practices that are used at SEP hatcheries and DFO affiliated facilities to promote fish health. These are all "acceptable practices" but may not all be used under all conditions or for all species. SEP encourages innovation and flexibility in fish culture operations to ensure the best possible treatment for the fish while at the same time considering operational constraints.

The following SOP's should be modified to reflect site specific practices that are for procedural reference and may be used for training purposes, while at the same time providing a framework to build "best practices" on.

Broodstock & Spawning

Broodstock represent an important and sensitive life stage. Fish are channeling their energy stores into the maturation of gametes while simultaneously undergoing the physical stresses related to migration, changing temperatures and re-entry into freshwater. The cumulative effects of these multiple stressors can result in a compromised immune system which can lead to ingress or reactivation of infectious agents. Failure to adequately address these concerns through proper husbandry techniques and appropriate biosecurity may lead to the introduction of pathogens into progeny or other fish on a facility and may potentially result in epidemics.

A female fish, heavily laden with eggs, cannot withstand the rough handling sometimes associated with poor hatchery practices. Great care should be taken during the sorting and spawning operation to dip up only two or three fish at a time. Never make a pass through a pen of nearly mature females and fill the bag of the dip net with fish. This can result in broken eggs, poor fertilization, and possibly permanent injury to the fish's reproductive system which is of concern with repeat spawners such as steelhead. It may safely be said that the less the fish are handled the better, and streamlining spawn-taking operations to reduce handling is certainly a step in the right direction.

The interval at which brood females should be sorted during the spawning season depends to a large extent on water temperature and season. To produce eggs of the best quality, it is necessary to watch the brood stock closely. The correct degree of ripeness must be attained in the females. Taking eggs before they are fully mature is as bad as not sorting frequently enough, which may allow some of the females to over ripen. If they are not sorted often enough, overripe eggs are sure to be found.

Research has shown that the ripening of eggs can be represented graphically as a curve with a sharp apex. The peak of this curve represents the time of optimum fertility of a particular lot of eggs, which must be stripped at that time. If taken prior to this date, lower fertility results, due to the eggs not being completely ripe. If taken later, on the down side of the curve, overripe eggs are encountered. Correct timing, through proper and frequent sorting, is one of the greatest secrets of successful egg taking.

In general, the size of the egg depends upon the size and age of the parent fish, the larger specimens producing more and larger eggs. Egg size also varies among different strains and

stocks of broodstock. It is reasonable to assume that competition among fry gives the larger fry a better chance for survival and faster growth. Size, however, can be attained only at the expense of number. There is, therefore, some point at which, on the average, the forces favoring size are balanced by those favoring number. The number of eggs produced by females of the same age and strain varies considerably.

The amount of sperm extruded from a male varies from a few drops to a teaspoonful. It has been stated that one drop of sperm will contain enough spermatozoa to fertilize 10,000 eggs. It is, of course, necessary for contact between sperm and eggs to occur; hence, the necessity of stirring the eggs and sperm together.

Since there is a limit in the time that both the eggs and the sperm remain viable, correct timing in the spawn taking operation is important. The length of time either eggs or sperm remain viable varies considerably and depends, perhaps, on several factors. Certainly, variety of fish and temperature are contributing factors. It is generally accepted that exposure of eggs or sperm to water for three minutes or more prior to fertilization will result in virtually a complete loss of viability. When eggs are broken in the spawn-taking operation, the process of fertilization is greatly hampered and at times completely stopped. Broken eggs in the spawning pan will appear as a white, creamy substance somewhat resembling sperm. This is actually the albumen from the broken eggs and, unless it is washed off immediately, some of it will lodge over the micropyle and present the spermatozoa from entering. Broken eggs probably contribute as much to poor fertilization as does any other factor. When albumen appears in the spawning pan, it should be washed off immediately, the sperm added, and the pan emptied of eggs before more are added.

4.1 Broodstock Selection

Rationale: Broodstock are selected to ensure that enhanced fish maintain the fitness characteristics of the native stock. This SOP addresses section *2.2.9* of the General Principles of Fish Health Management. The goal of this SOP is to ensure the selection of broodstock that maintain the fitness characteristics of the native stock.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

The percentage of Jacks in the broodstock should reflect the proportion in the population.

Some captured adults may be released to achieve a balanced sex or age ratio or to exclude any fish with apparently questionable health status.

All attempts should be made to ensure the selected broodstock population contains individuals collected across the time course (early, middle and late) of the run.

Details of the Operating Procedure:

Nitinat Hatchery does not use a brailer system for collecting and moving fish. Fish either swim into the holding ponds via the fishway, or are collected offsite by seine net. Fish in the adult holding ponds and/or the lake net pens are crowded and moved into sorting using a pescalator.

All fish are selected in a random manner and jacks are not selected against.

Broodstock will be rejected if fish display obvious external signs of disease such as ulcers.

4.1.1 *Chinook*

The majority of Chinook are collected from offsite as they are difficult to attract into the fishway. 70-80% of the Nitinat Chinook and 100% of the Sarita Chinook are collected offsite by seine net. Generally these fish are collected by beach seine using a jet boat to pull the net out and around the school of fish. If this method is unsuccessful, purse seining may be used as an alternative collection method.

An indicator pool will be used for first set analysis. All fish, including those not retained for brood, are counted. This provides an indication of the numbers of fish in the river. Sets will be repeated 2-3 times to generate approximately 80% of the required fish for brood.

Sarita Chinook are transported at half the density that the Nitinat Chinook are transported at.

Fish are only rejected as brood stock if they are overly ripe or if one sex is over-represented in the collection.

Notes:

- Each days set will be in a different location
- Once the fish are in the adult holding ponds, regular sorting for sex and degree of ripeness will be carried out. Marks will be recorded at this time
- Sorts are carried out through the use of a pescalator (See Broodstock Handling)
- At least half of the required broodstock are collected in the first set
- Approximately 500 adults can be held in each holding pond
- The first set is determined by historical timing and generally is carried out during the third week of September. Swim surveys will provide information on where the fish are localizing the week prior to the planned set
- Chinook egg takes are generally completed by the middle of October with the peak population in the river around the 10th. By this time 80-90% of the eggs required will have been taken. If there are still large numbers of fish entering the system after this period, additional egg takes may be performed to represent the late genetic stock, but generally these will be surplused to processing

4.1.2 *Coho*

Coho are predominantly collected as swim-ins. Because a minimum of 10=20% are required to be wild fish, some beach seining will be required.

Some early coho may be taken as broodstock and held so that a wider component of the run is represented. Collection of unmarked fish will begin in early September and may coincide with Chinook collection. These fish will be held for ripening. Depending on fish condition, they may simply be counted and released or they may be sorted and unmarked adults in good condition collected and held in the circular tubs.

Fish will be selected across the entire run timing regardless of whether they are swim-ins of seined.

4.1.3 *Chum*

Chum are collected both through seining and as swim-ins dependent on conditions. Adults generally enter the system in a ripe state and are processed quickly.

4.1.4 *Sorting*

Nitinat Hatchery does not use a brailer. A pescalator is used to move fish from the ponds into the sorting system. The sorting is outlined under Broodstock Handling

Forms & Records:
Sort sheets
Adult capture/capture and transporting form
Adult holding/brailer activity form
Weigh ticket form
Tote sort form

Escapement summary form (mark sampling)
EnPro


Duksbak adult sheets


Biosampling Form


Stream Survery Form


SURPLUS.XLS


TOTESORT.XLS

4.2 Broodstock Handling

Rationale: Broodstock will need to be handled at least once to assess gender and degree of 'ripeness'. Fish must be handled with care to protect the brood fish and subsequent gamete quality. This SOP addresses section *2.2.9* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that fish are handled with care and subjected to minimal stress.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Fish may be crowded to make brailing or dip netting easier. The duration of crowding and the density should be kept to the minimum amount of time possible. Nets used for crowding and netting should be knotless and numbers of fish in the nets should be kept low (<1/3 of the net volume) to prevent fish on the bottom of the load from being crushed.

If using anaesthetics (see *Anaesthesia* SOP), the fish should be monitored until they are ready to be handled. The anaesthetic bath may contain mucus protectants (e.g. Vidalife™) to protect the fish's cuticle from subsequent opportunistic infection. Water quality should be monitored during anaesthesia. When easily handled, fish should be netted out of the anaesthetic bath and assessed for 'ripeness'. If antibiotics have been prescribed by the Veterinarian (see *Broodstock Treatments* SOP), they may be injected at this point.

Female fish are considered ripe when the body wall feels soft and thin and loose eggs are palpable within the coelomic cavity. Male fish are considered ripe when milt is easily expressed. Milt should be white and opaque; if the fluid is clear or watery, the fish is not yet ripe.

Fish are handled with care to minimize scale and mucus loss and are not held solely by the tail if expected to survive post-handling. After any anaesthesia and handling event, fish are monitored closely for signs of injury, morbidity and mortality.

Details of the Operating Procedure:

4.2.1 *Beach Seining for Chinook*

Adult Chinook may be seined by beach seine or by purse seine. Fish may be collected and transported to the hatchery for holding, or they may be held in lake net pens and spawned at the lake site.

Equipment checklist

Crew (enough to do the job)	Clubs
Equipment for the crew (raingear, etc.)	Twirl packs
Mitts/gloves	Kitchen catcher
Boat and motor	Spawning knife

Gas for outboard	Sampling gear (see sampling checklist)
Net	Icepack
Buckets	Foam pad
Tarp	Toolkits
Umbrella	Lifejackets (for everyone)
Dipnets	Net mending apparatus (twine, needles)
Cooler equipment	

1. Prepare all equipment
2. Ensure that the three transport trucks are available and prepared
3. Fill the transport tank with water from the aeration tower
4. Set the seine net and haul it in until the fish are crowded
5. Keeping a rough count of the ratio of males to females, sort the fish in the net by hand (wearing gloves)
6. Working in teams of two, attach a tailer to each fish and, keeping the fish in the water, transport it to the beach
7. Pass the fish to a staff member on the beach ensuring that the body is supported and that the fish is not carried solely by the tailer
8. Cradling the fish carry it to the transport tank and pass it to the staff member on the transport truck for placement inside the tank
9. Remove the tailer on release into the transport tank
10. When the desired density has been reached, close the lid on the tank securely and transport the fish to the hatchery for release into the ponds
11. Record the number of fish placed into each holding pond

4.2.2 *Adult Capture from Nitinat River*

Staff Required: Minimum - 12 Maximum - 16

Transporters	3 – 4
Hatchery observer	1
Tailers	2 – 3
Packers	4 – 6
Loaders	1
Data recorder	1

1. Set up the fish ladder, brail pool, release way & Ponds 9, 10, 11 & 12
2. Set the pond flows at 7000 L/min
3. Ensure that the pond covers are in place on ponds 9 & 12
4. Install 3 transport tanks [small] on hatchery ¾ ton trucks & the large transport tank on the 1 ton flat deck

5. Set up the 150hp jet boat with the VHF radio, life jackets, boat kit, fuel & seine net.
6. Ensure tailers, whirl-packs, egg collection bags, fish handling mitts &gloves, tarps, rope bleeding knife, tally counters, water proof note books, pencils, freezer packs & cooler and any other required equipment is ready for use
7. Ensure that a discussion has been had with all staff involved stressing safety and accuracy of counts
8. Inspect the collection pool to verify fish abundance and determine how large a set should be made (taking into account crew size, water quality and temperature).
9. Delegate two people to the boat to make the set
10. Attach the boat to the corks on the outside of the set
11. Slowly work both ends of the seine net bring the net to shore with the lead line preceding the corks.
12. If the fish are displaying indications of stress stop crowding and let them settle down.
13. Tail adults and them passed to the packers
14. The packers transfer the fish to the waiting truck where they are counted in and sexed
15. Ensure that the number of fish collected is accurately recorded by the data collection person.
16. All non targeted species are sexed and released over the corks with all the number & sex of each species recorded.
17. Decide if eggs will be collected from ripe females at the river site
18. When the set is empty reload the net on the boat
19. Collect all tailers and other equipment
20. Return to the hatchery and prepare for the next day

4.2.3 ***Using the Adult handling System***

1. Set up the pescalator
2. Set up the sorting table
3. Set up the egg take shelter and bleeding racks/tubes etc
4. Set up the anaesthetic (clove oil) and euthanasia (CO_2) baths (see Anaesthesia)
    a. Turn off the drain valves to the anaesthetic tank, the 2 kill tanks, the sorting table & all drain valve lines
    b. Add 135ml of Eugenol solution to the anaesthetic tank
    c. Fill the anaesthetic tank using the wash down hose from the brail pool standpipe
    d. Fill the kill tanks from the standpipe at the hydraulic pump
    e. Connect the water lines that feed adult return lines and turn them on
    f. Turn on the standpipe line feeding the female table sprinklers
    g. Charge the kill tanks with CO_2 at 40 L/min (full flow) for 15 min then reduce

h. Turn on the hydraulic pump and the pescalator (check to ensure that the anaesthetic flow through the pescalator is 3 L /min (each revolution of the screw takes 20 s)

i. Turn on the anaesthetic pump to deliver to the pescalator

j. Crowd adults to pescalator intake

k. Adjust speed and anaesthetic flow to ensure adults are “knocked out” sufficiently to handle

l. At the end of the sorting period (noon) drain the anaesthetic tank and refill.

5. Fill the male and female kill tanks with water and add carbon-dioxide
6. When water has reached saturation reduce the flow
7. Fill the round tank with water and add eugenol mixed with ethanol
8. Add sodium bi-carbonate to the tanks as a buffer
9. Start the hydraulic system and allow it to warm up.
10. Set up the crowding channel (release-way)
11. Install the attraction channel stop logs
12. Turn on the appropriate wells
13. Adjust the pond flows
14. Ensure the channel crowder is pulled to the end of the crowding channel (the end opposite the pescalator)
15. Lower the water level in the release-way and the pond scheduled for sorting by removing the finger weir and stop logs
16. Crowd the appropriate # of fish into the crowding channel below the pond (don’t overfill the channel)
17. When an appropriate number of fish are in the crowding channel replace the outflow screen at the outflow of the pond to stop fish from entering back into the pond
18. Start the fish screw and adjust the rotation to 2.5 rotations per/min (speed will be adjusted relative to the size of the fish and relative to the use of anaesthetic (slower if using anaesthetic). When the fish enter the pescalator, they are dewatered via screens

    a. Anaesthetic (Clove oil) is recirculated through the pescalator and is refreshed approximately every two hours

    b. The pescalator takes approximately 45-60 seconds to lift a fish from the channel to the top and onto the sort table. Therefore, the anaesthetic is sufficient to sedate them enough to handle for ripeness checks

19. Activate the channel crowder to crowd fish into the pescalator.

    a. Do not overcrowd fish or they will be overly stressed the pescalator will become overloaded and eggs will be lost

    b. Generally, it is best to crowd up and compress the fish and run the pescalator until the pressure is alleviated.

c. At that point move the channel crowder forward again to compress the fish once more.

d. Repeat this process until the fish in the crowding channel have been removed or until the sort has met the egg take requirements for the day.

20. Fish are directed down the chute to the sorters who identify which fish are to be used for egg and milt take. Fish are sorted by sex and by ripeness. Sorts and ripeness checks are to be performed by experienced staff
21. When the adults arrive on the table sort them for ripeness
22. Sort the ripe males and females into their respective kill tanks
23. All green adults are returned to a holding pond (the upper 1/3 section) or sent to surplus
24. If the adults are too active on the sorting table slow down the rotation of the screw
25. If they are too submissive speed up the rotation to allow easy checking as well as a speedy recovery of the adults sent back to the holding pond
26. When the adults in the kill tanks have succumbed, rotate the finger paddles and allow the adults to go onto the spawning tables
27. Slash the gills of the female fish and allow them to bleed out prior to spawning
28. NOTE ALL ADULTS HANDLED ON THE SPAWNING TABLES MUST BE CHECKED FOR MARKS PRIOR TO DISCARDING. ALL MARKED ADULTS ARE TO BE PLACED IN THE SAMPLING TOTE
29. Females will be held for the egg-taker in a method that prevents sun or water from entering the egg bucket
30. When the pond is empty or the release way is full replace the stop logs

4.2.3.1 Females

1. Feel each fish for ripeness while holding it in an upright/horizontal position
2. A small amount of pressure on the belly will cause some eggs to squirt out and this is the sign of a ripe fish
3. If the fish is ripe, apply a tailer
4. Holding it by the tailer with the tail slightly elevated and with the head laying on the ground, kill the fish with several sharp blows to the head, between the eyes, using a small aluminum bat
5. If the fish is green fish, place it into the “sorted” pond for the designated species or send to surplus
6. Recheck and resort in 5-7 days

4.2.3.2 Males

1. Remove males from the sort table and hold them upright horizontally (same as females)
2. Run hand down belly wall towards the vent starting at the head of the fish, squeezing gently with moderate pressure, if sperm is ejected, the fish is ripe

3. If the fish is green fish, place it into the “sorted” pond for the designated species
4. Recheck and re-sort in 5-7 days

4.2.4 ***Ripe fish***

1. Direct fish to be used for gamete collection into the CO_2 euthanasia tank
2. When the fish have succumbed to the CO_2, transfer them to the outlet table
3. Cut the gills using a sharp hooked knife
4. Push the fish to the runoff table and allow them to bleed on the bleed out ramp – fish are not clubbed
5. When the fish have been bled, turn on the sprinkler heads to rinse the fish of and remove excess blood and contaminants
6. Slide the fish down to the gamete takers, males are sent down one chute, females down another

Notes:

- If green fish come through to the egg taker – they are noted
- All fish are counted and sex, marks, species, etc are noted on the tally sheet. The tallies will be verified by the inside records

At the end of the day …….

- Turn off hydraulics and anaesthetic pump
- Turn off standpipes to the kill tanks after they have been drained, cleaned and refilled for the next day (Do not charge)
- Make sure all drain valves on the female table line have been opened to avoid freezing and the splitting of the line.
- Wash down work area
- Clean up equipment and put away
- Ensure data is correct and handed in

4.2.5 ***Sarita Chinook***

Sarita Chinook are brought to the hatchery from another watershed and are therefore held separately. These fish are held on well water and a cover is placed over the water top reduce stress and predation. These fish are not treated with formalin and are not processed using the pescalator. Fish are crowded manually and dip netted individually into a TMS anaesthetic bath for determination of ripeness. The holding pond is shallow so the levels do not need to be reduced to enable collection.

4.2.6 ***Adult Capture from Sarita River***

Staff Required: Minimum - 11 Maximum - 14

Transporters	3 – 4
Hatchery observer	1
Tailers	2
Packers & Loaders	4 – 6
Data recorder	1

Set-up:

1. Set up the release-way & Pond 5 & Ponds 9, 10, 11 & 12
2. Set the pond flows to 3100 L/min
3. Ensure that the pond covers are in place on the ponds
4. Install 3 transport tanks [small] on hatchery ¾ ton trucks & the large transport tank on the 1 ton flat deck
5. Call the Huu-ay-aht Band and ask them to swim the river to find brood stock and their locations
6. Install the Dura boat and 9.9 outboard on the trailer and ensure that the required equipment is aboard (life jackets, boat kit, fuel & Sarita seine net)
7. Prepare: tailors, whirl-paks, egg collection bags, fish handling mitts &gloves, tarps, rope bleeding knife, tally counters, water proof note books, pencils, freezer packs & cooler

Action:

1. Inform the Huu-ay-aht Band of the time and place where brood stock collection is planned
2. Ensure that a discussion has been had with all staff involved stressing safety and the accuracy of counts
3. Inspect the collection pool to verify fish abundance and determine how large a set should be made (taking into account crew size, water quality and temperature)
4. Have the Huu-ay-aht swimmer swim the pool to verify abundance & species
5. Delegate two people to the boat to make the set
6. Attach the boat to the corks on the outside of the set
7. Slowly work both ends of the seine net bring the net to shore with the lead line preceding the corks.
8. If the fish are displaying indications of stress stop crowding and let them settle down
9. Firmly grasp each fish and turn it upside down to calm it
10. Place adults into holding bags and maintain the bags securely in a pool in the river until transporting begins. If the truck is en route, fish may be held temporarily in a holding pen set into the river
11. Take the holding bags to the waiting truck when they are enough fish to make a full load.

12. Count in the adults and sex them as they are loaded
13. Ensure that the number of fish collected is accurately recorded by the data collection person
14. Sex, record, and release all non-targeted species over the corks
15. Decide if eggs will be collected from ripe females at the river site
16. When the set is empty reload the net on the boat
17. Collect all tailers and other equipment
18. Return to the hatchery and prepare for the next day

Procedure	***Hatchery***	***River***	***Lake***
Method of capture:	*Crowding swim-ins*	*Beach Seine*	*Seine*
Record species:	*Chinook, Chum, Coho, Steelhead or Cutthroat (or other Species)*		
Record sex:	*Male, Female, Jack, or Immature*		
Enumeration:	*Fish will be counted OUT of the pond.*	***All fish*** *caught in the 1st Set will be counted. Only fish killed in subsequent sets will be counted.*	*Fish transported to the Hatchery as broodstock will be counted OUT of the pond. Sampling will be done to obtain accurate numbers of fish disposed of in totes.*
Sort fish when caught:	*All fish will be sorted and counted by sex*	*Kill only ripe females and corresponding males/jacks.*	*Hold females until ripe at lake or transport to Hatchery*
Check for Marks!! *Where possible, double checks for marks will be made.*	***All swim-in fish*** *will be checked for marks; marked fish will be disposed of by hatchery (not sold)*	***Only fish killed on the river*** *to be sampled. Any discarded or released fish (unripe females, excess males/jacks) will NOT be sampled for marks; additional sets in a similar pool or location will not be counted by sex; only broodstock or morts will be counted and checked for marks*	***As many totes as possible from the lake*** *will be checked for marks; marked fish will be disposed of by hatchery (not sold)*
Fish Sales: *Hatchery staff will determine grade quality.*	*Sold as either Hatchery or Carcass grade; all female broodstock will be sold as carcass.*	*If sale fish are desired by the fish buyer, it is his/her responsibility for the retrieval of these fish from the river, unless a prior agreement is made.*	*Sold as either Hatchery or Carcass grade; all female broodstock will be sold as carcass; Numbers/sex of fish per tote will be recorded; all totes will be labelled*
Record Keeping requirements:	*Date, species, location, (set), initials, and all pertinent information, including number of fish caught, checked for marks, sampled, spawned, sold, disposed of, or released.*		

4.2.7 *Adult Holding in Concrete Raceways*

Note: all Chinook held in deep or shallow ponds must have a pond cover in place!

Flows for Chinook are to be set at a minimum of 1 litre of water/kg of fish/min

Weight /kg	Species	Pond #'s	Loadings	Min. Flow in L/min	Water Depth
11.5	Chinook	9 - 12	600 – 650	7000 - 7500	1.2m or 4'
11.5	Chinook	1 - 8	225 – 270	2900 - 3100	0.5m or 1.5'

Flows for chum are to be set at a minimum of 0.5 litre of water / kg of fish /min

Weight /kg	Species	Pond #'s	Loadings	Min. Flow in lpm.	Water Depth
7.0	Chum	9 - 12	1600 – 2000	5600 - 7000	1.2m or 4'
7.0	Chum	1 - 8	1200 – 1500	4200 - 5250	0.5m or 1.5'

Note: When adults are being held in concrete raceways exterior lighting located in the following areas must be turned off:

- The lights at the south-end of buildings 1 – 2 & 3.
- All access lights on the east and west sides along the southern end of buildings 1 – 2 & 3 .

1. No excessive activity must be carried out after hours that may stress the holding adults
2. Stand-by personnel should check on their final rounds to ensure covers are in place and lights are turned off

					Holding Capacity	
Location	**#**	**Description**	**Volume**	**Load Rate**	**Chinook**	**Chum**
Hatchery	4	Deep Ponds 9 - 12	225 m3	1 Kg/ lpm	900	2100
Hatchery	8	Shallow Ponds 1 - 8	122 m3		500	1400
Lake		Netpen (40 x 40 x 20)	864 m2	Good flow	N/A	4000 max
Lake		Netpen (20 x 40 x 20)	432 m2		N/A	2000 max

Forms & Records:

References:

Broodstock Treatments
Anaesthesia
Fish Handling Procedures

Adult Handling STEPS

4.3 Broodstock Biosecurity

Rationale: Broodstock represent a sensitive life stage. They are more susceptible to pathogens that they may be carrying or to which they are exposed due to physiological changes associated with maturation. It is important to protect broodstock and their gametes from infectious disease-causing agents. This SOP addresses section *2.2.9* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that broodstock are protected from pathogens and that other fish groups are protected from pathogens that broodstock may be carrying.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Broodstock should be maintained in a separate holding area from other fish (i.e. juveniles). All broodstock holding areas should be pressure washed and scrubbed and disinfected with bleach, or other suitable disinfectant, prior to being used for other life stages.

All equipment used on broodstock should be designated for brood use only. Staff separation should occur whenever possible.

Disinfectant foot bath stations should be regularly maintained between adult holding and incubation areas. Spray bottles of Ovadine™, or other topical disinfectant solution, should be available for surface disinfection of hands and rain gear.

Staff must adhere closely to site and staff biosecurity procedures (see *Site and staff disinfection and biosecurity* SOP).

Details of the Operating Procedure:

The adult holding ponds are surrounded by chain link fencing that is electrified to keep bears out.

Formalin may be used as a prophylactic treatment to keep fungus under control during holding.

Sarita Chinook are brought from another watershed and are therefore held separately. These fish are held on well water and a cover is placed over the water top reduce stress and predation.

Separate equipment (nets, brushes etc) is used for broodstock to reduce pathogen transfer potential. Separate equipment is used for different stocks.

Vidalife is added to the transport water to reduce stress levels. Vidalife is used according to manufacturer's directions. Ice may be added to transport water for Sarita Chinook.

Other stocks, try to keep densities down, spread the fish out evenly in the ponds, sort swim-ins first (at the lower end of the ponds, oxygen injection system.

Sex sorts are performed on a weekly basis, or more often if required. Sarita stocks are sorted by hand rather than through the use of the pescalator.

Cross contamination is reduced during spawning by doing egg takes on different stocks on different days or at a different time of day. Egg takes are not run on multiple stocks simultaneously.

Different stocks are physically separated on site.

The numbers of chum in the facility tend to increase rapidly. Sorting is as frequent as possible to keep densities under control. If necessary, additional oxygen may be delivered via the oxygenation system.

Forms & Records:
Inventory and location records

References:
Site and staff disinfection and biosecurity


Nielson fish pump STEP


Fish Pesculator (Lake) STEPS

4.4 Adult Carcass Disposal

Rationale: Carcasses should be disposed of in a manner that minimizes the potential for spread of disease. This SOP addresses section *2.3.3.2* of the General Principles of Fish Health Management. The goal of this SOP is to ensure disposal of carcasses consistent with a manner to lower the possible spread of disease agents.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Potential methods for carcass disposal include:

- Carcasses may be frozen and disposed of into solid waste containers and eventually removed to municipal landfill. Incineration and composting are also possible alternatives.

- Carcasses may be buried on site as a facility allows, however, this entails a risk of predator attraction. Alternating layers with sawdust and lime may be needed

- Fish that are surplus to brood requirements may be harvested for commercial sale, providing they have no potential drug residues in their tissues (i.e. no anaesthetics have been used during their harvest).

- Carcasses may be placed into their natal streams to provide nutrient enrichment according to the DFO Carcass Placement Guidelines. This use is permitted under the authority of the intergovernmental Introductions and Transfers Committee and is not suitable for all facilities

- *Because of drug clearance times, and the length of holding, fish previously treated with an antibiotic or anaesthetic must not be used for carcass placement. However, fish treated with external chemicals that do not require a withdrawal period (e.g. Parasite S or Chloramine T) are considered safe for placement. If in doubt, contact the Fish Pathology Program at PBS.*

Details of the Operating Procedure:

Morts are picked from ponds on a daily basis. The sex, number, species etc, are recorded.

Spawned out carcasses are placed in totes and moved aside. Carcasses are placed into a wheelbarrow before being counted and sexed. Any marks are noted and recorded. Carcasses are generally placed into totes and removed by the local First Nations for use as ceremonial food fish, for planting in the river, or for reduction. If they are not sent to the reduction plant, they are planted into the Little Nitinat River.

Forms & Records:
Records of numbers and species of carcasses placed in streams are kept.


Native Food Fish Slip.doc


Nitinat Fish Sale Weigh Slip.doc


Overall Fish Slip.doc

References:

Guidelines for in-Stream Placement of Salmon Carcasses for Nutrient Enrichment

4.5 Gamete Collection (Egg Take and Milt Collection)

Rationale: Attention to hygiene at egg take will decrease the risk of horizontal pathogen transfer to other brood fish or progeny. This SOP addresses section *2.1.3.6* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that gamete collection is performed in as hygienic a manner as possible.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Procedures:

4.5.1 ***Prior to gamete collection***

All necessary equipment should be cleaned, disinfected and dried before and after use.

All necessary transport/transfer permits should be in place.

In the event of disease screening, Lab personnel should be contacted to ensure they know of the workload on the way and to avoid conflicts and shortage of assistance.

4.5.2 ***Female fish:***

Ripe females should be euthanized by a sharp blow to the head. The gill arches should be cut and the fish hung by the tail to bleed for 5 – 10 minutes. Blood transfer to the surrounding area may be minimized by placing paper towel inside the operculum.

Where broodstock disease screening, specific pairings or matrix spawning is planned, each fish should be assigned a number. The number should also be written on the bag into which the eggs will be collected. It is critical to track progeny location to allow for destruction of the gametes of pathogen-positive brood fish.

The fish should be handled in an inverted position until it is ready to be incised for egg removal.

After bleeding, the ventral surface of the fish should be wiped down with paper towel to minimize blood, water or mucus dropping into the eggs. A disinfectant solution may be applied to the external surface of the fish as well, to reduce the risk of vertical transmission of disease.

The person cutting the fish should clean their hands or change to a new pair of gloves between females. A new spawning knife should be used for each female or the spawning knife should be disinfected between uses on different fish (see *Equipment disinfection*).

The fish should be turned into a head up position by the handler and the person cutting the fish cuts from the vent towards the head, cutting to the side of the pelvic girdle.

The eggs should be collected into a labeled, clean, disinfected and dried bucket.

Any eggs with abnormal appearance, cloudy ovarian fluid or from a female with obvious signs of disease should be immediately discarded.

Eggs may be topped with oxygen and should be placed into a cool environment (e.g. on gel packs covered with paper towel or newspaper) until fertilization or transport. The eggs should not sit directly on the ice or gel pack, a layer of newspaper should be placed in between.

Samples may be collected from the female carcass for disease screening.

The person responsible for stripping the eggs from the female should wash his/her hands or change gloves and spawning knife prior to handling another female.

4.5.3 ***Male Fish:***

Male fish may or may not be euthanized for milt collection. When they are used for multiple collections, careful handling is essential and the fish should be returned to the holding unit and monitored for recovery after collection.

Ripe males may be euthanized by a sharp blow to the head. If milt collection will be repeated, the male should be anaesthetized, handled expediently and monitored during recovery.

Each fish may be assigned a number. Where fish are used for multiple collections of milt they should be clearly identifiable (e.g. pit tagged). A Whirl-pak™ bag may be labeled with the fish's number.

The ventral body wall should be wiped down with a clean paper towel to minimize water or mucus dripping into the milt being collected. A surface disinfectant may be applied to the external surface of the fish to reduce the risk of vertical transmission of disease.

Milt is collected by cradling the fish, extending the tail while applying firm but gentle pressure on both sides of the body wall at the level of the testes. Milt should be collected into a sterile Whirl-pak™ bag or cup.

Any milt with abnormal appearance, or containing blood or feces, should be immediately discarded.

Milt may be stored in a plastic container topped with oxygen. The surface area of milt exposed to oxygen should be maximized. The containers of milt should be put into a cool environment (i.e. onto ice packs covered with newspaper or paper towel to maintain a cool temperature).

Milt may be checked for motility prior to use (a drop of milt plus a drop of water for sperm activation, examined under magnification within 10 seconds).

Details of the Operating Procedure:

4.5.4 ***General Spawning Notes***

- Poor quality sperm (coagulated, excessively watery, containing bile or faeces etc.) will be discarded in incubation
- Any females that are found to contain bloody fluid, green eggs, bloody eggsm water hardened or ruptured eggs, will not be used for brood
- Make sure eggs and sperm have plenty of air in their respective bags/whirlpacks, and that they are kept cool, dry and out of sunlight/rain
- Every morning the fertility of the eggs from the egg take of the previous day will be checked. One hundred random eggs will be sampled from these. All data will be recorded and passed on to the S.E.O. and the A.S.E.O. to be checked and any necessary changes can be made.

Spawning Procedure	**Chinook**	**Sarita CK**	**Chum**	**Coho**
Spawn Ratio (F:M)	1 : 1	1 : 1 *Matrix*	5 : 2	1 : 2 *Matrix*
Broodstock selection	Usual health checks	Usual health checks	Usual health checks	*Test* for **BKD**
Bleed/Hang	Yes	Yes	No; unless time/people	Yes
Bags ("Glad Kitchen Catchers")	To help combat contaminants, the use of bags to contain and transport eggs to incubation can be used. For Coho and Sarita Chinook, use 1 bag per female.			
Transporting	Transport eggs and sperm to hatchery ASAP.			

Egg Take Equipment Checklist

- □ *Clubs*
- □ *spawning knives*
- □ *buckets*
- □ *lids for buckets*
- □ *spawning bench*
- □ *dip net(s)*
- □ *ice (if warm)*
- □ *insulated tote with lid*
- □ *glad kitchen catchers*
- □ *gloves*
- □ *bleeding knife*
- □ *tailers*
- □ *spawning basin*
- □ *paper towels*
- □ *holding hooks*
- □ *cooler*
- □ *sperm containers with lids or Whirl-pak bags*

4.5.5 ***Staff required:***

Chinook: Minimum- 9 Maximum-11

Sorter	1
Bleeder	1
Sperm collection	2 – 3
Egg taker	1
Holders	1 – 2
Carcass & sampling	1
Incubation	2 - 3

Note: incubation personnel will be helping to take eggs and sperm when there are no buckets of un-planted eggs present in the incubation room

Chum: Minimum- 10 Maximum-15

Sorter	1
Bleeder	1
Sperm collection	2 – 3
Egg taker	1 - 2
Holders	2 – 4
Carcass & sampling	1
Incubation	2 - 3

Note: Incubation personnel will be helping to take eggs and sperm when there are no buckets of un-planted eggs present in the incubation room. Their normal duties involve weighing, sampling, fertilizing, sampling and planting eggs as well as recording all required data and cleaning buckets for reuse at the egg take station.

4.5.6 ***Gamete Collection Procedure***

1. Ensure that egg take buckets have been precleaned, predisinfected and rinsed in a sodium bicarbonate solution

Females

2. The egg taker will remove an egg take bucket from the stack and set it on the ground in front of him/her
3. The assistant removes a female fish from the table, keeping the tail elevated, and presents it to the egg taker
4. When the egg taker is ready to remove the eggs, drop the tail of the fish down as the egg taker cuts the ventral wall, this will allow the eggs to spill out into the egg take bucket
5. When stripping eggs, take care not to break any eggs; do not force eggs from the skein - take those that fall away with a gentle shake

6. Check females for disease (kidney lesions, boils, hemorrhaging, cloudy ovarian fluid). Any female that is suspected of poor health, or unhealthy or water hardened eggs will be discarded. If eggs from an unhealthy female have been put in a bucket with healthy eggs, all the eggs in the bag are discarded
7. After spawning place the carcass into the removal chute and send them to a fish tote

Males

1. Males are killed in the same manner as for females (above) but not bled prior to stripping
2. Jacks are noted but not selected against
3. The handler removes a ripe male from the sort table and turns towards the assistant
4. Place a foot on an upturned bucket and support the male fish on the knee
5. Check the flow of milt by stripping a small amount to the side
6. If the milt appears of good quality, move a bucket of eggs into place and strip a quantity of milt into the bucket

Note: In the morning, prior to the egg take, express milt from 20-40 males into individual whirl-pak bags and place them onto wrapped ice packs in a cooler. This provides a reserve in case the numbers are incorrect at the end of the day. Ensure that no sun or water gets in the bag. When the milt is sent to the incubation room a paper with the # of males and jacks is to accompany the sperm

7. If a male does not provide an adequate amount of sperm, another may be expressed into the bucket of eggs
8. After spawning place the carcass into the removal chute and send them to a fish tote

Transfer to Incubation

1. Once buckets have been filled with eggs and milt has been expressed into them, they are gently mixed by hand
2. When the pre-determined number of females have been stripped into the bucket and the milt has been added, place the lid on top
3. Place the buckets on the roller for transfer to the golf cart (16 buckets will fit in the back and 4 in the front)
4. When 20 buckets have been filled, place a tally sheet in the first bucket (indicates number of females in the 20 buckets) and transfer them to the conveyor rack to send them to the incubation room
5. Clean and disinfect all buckets, rinse them with a sodium bicarbonate solution and stack them upside down

Note: The Sarita Chinook program uses a separate set of equipment. Sarita eggs are taken into buckets lined with plastic bags. Milt is taken in Whirl-pak bags. Gametes are mixed in the incubation room rather than at the egg take shelter. Males may be re-used depending on available numbers. If males are to be re-used, clip the adipose fin to identify them as having been stripped once. The egg wash water is directed into a disinfectant tote and all equipment is disinfected in an Ovadine bath.


4.5.7 ***Species/Stock specific notes***

4.5.7.1 Coho

Following gamete collection, kidney tissue is removed (according to the BKD sampling procedure (revised 2006)) for submission to the pathology lab at the Pacific Biological Station.

4.5.7.2 Chinook

<u>Field</u>

- All females killed in the field eggs will be bled prior to taking of the eggs.
- For every female killed, one male will be killed also.

  *Note: Jacks constitute a male killed
- All eggs will be taken into "Glad Kitchen Catcher" bags with one female for each bag. The bag will be tied with air entrapped and will then be placed in a bucket and kept cool. They will be returned to the Hatchery within the hour.
- All sperm will be collected in Whirl-Paks as is done with Hatchery egg take.
- All salmon from the first set from each area are to be counted and kept separate as to specie and sex. All Jacks are to be kept separate.

Hatchery

- All females used for eggs will be bled.
- All eggs will be discarded from any female having cloudy ovarian fluid or water hardened eggs.
- No bags will be used for the eggs. The eggs from each female will be put in a separate bucket then taken to the incubation room for fertilizing and planting.
- Males will be killed after checking for sperm availability.
- Sperm will be taken in Whirl-paks and then transported to the incubation room for the fertilizing and planting procedure.

*Note: Sperm is to be kept cool and out of the direct sunlight or rain.

4.5.7.3 Chum

Field

- Chum are collected by seine net
- No bleeding of chum is required before spawning.
- For every female killed, one male will also be killed.
- All eggs will be taken in "Glad Kitchen Catchers" with the eggs from five females put in each bag. The bag will be tied with air entrapped, placed in a bucket and kept cool. They will be transported to the Hatchery within an hour.
- All sperm will be collected in Whirl packs as per Hatchery operations.
- All salmon are to be counted from the first set taken from each area, and kept separate as to specie and sex.

Hatchery

- Bleeding of females is not required, but may be done if time or manpower permits.
- All eggs will be discarded if taken from a female with cloudy ovarian fluid or with water hardened eggs.
- No bags will be used for eggs at the Hatchery unless there is a shortage of buckets.
- All buckets will be rinsed from the overflow of the boxes or cells, and the excess water either wiped off, or shaken off prior to putting eggs into them.
- Males will be killed at the start of every egg take day, and their milt collected in Whirl packs.

*Note: Sperm is to be kept cool and out of the direct sunlight or rain.

Forms & Records:

Egg Take Forms

Spawn Sheet.xls

Number Spawned Tags

References*:*

Equipment disinfection
Anaesthesia
Diagnostic Sampling protocols
Broodstock Handling
Egg and Milt Transport

Bicarbonate rinsing

4.6 Egg and Milt Transport

Rationale: Gametes must be transported properly to maintain their viability. Strict biosecurity protocols must be in place to minimize pathogen transfer from the broodstock location to the hatchery. The goal of this SOP is to ensure that gametes are transported safely to the hatchery and pathogen spread is minimized between the spawning site and the hatchery site.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Procedures:

All necessary permits/licences should be in place

If eyed eggs are being transferred under ITC permit to complete incubation at a new site, the eggs and transport water should be disinfected on arrival at the new site as stipulated by the transfer permit. (See the Federal-Provincial Introductions and Transfers Committee web page for further information)

If disease screening is planned, all gametes should be clearly labeled to identify the number of the doe or buck from which they originated. This is critical in ensuring that any offspring of pathogen-positive broodstock can be discarded later.

Eggs should be contained in clean plastic bags and may be topped with oxygen. The bags containing the gametes should be handled in a hygienic manner.

Milt may be contained in sterile Whirl-pak™ bags or small sterile styrofoam cups with lids. Containers may be topped up with oxygen. The amount of air space in the bag should be at least 2/3 of the bag volume and bags should lie on their sides to maximize fluid surface area exposure to oxygen.

Gametes should be placed into insulated containers and their temperature maintained as close to the originating temperature as possible. The containers should be clean, have secure lids, and be labeled if screening or matrix spawning is planned.

The outside of the container should be disinfected prior to transport, and site and staff biosecurity protocols are followed by the staff member transporting the eggs.

Details of the Operating Procedure:

Coho eggs are taken at the river collection site but male fish may be held at the hatchery. This is because difficulties have been encountered with sperm viability from fish at the river. ./When eggs are returned to the hatchery the appropriate number of males will be stripped for fertilizing the eggs. As fish are collected, the numbers are radioed to the hatchery to communicate the number of males required. 2 males are used to fertilize 5 females.

1. Collect eggs into plastic bags

2. Tie off the open end of the bag ensuring that the excess space is filled with air
3. Place the lid on the bucket
4. Stack the buckets in the back of the truck
5. It is generally cool during egg takes and ice is usually not required
6. Radio the hatchery staff to inform them of the number of females stripped so that they can strip the appropriate number of males for fertilization on arrival
7. When the eggs are planted in the incubators, mark the incubator as containing transport eggs

If Chinook are stripped offsite, fertilization is performed at the hatchery, not on the river.

1. Collect and transport eggs as described earlier.
2. Collect milt into Whirl-pak bags
3. Fill bags with 1/3 milt and 2/3 air to provide an adequate surface area for oxygen transfer
4. Place inside a cooler containing wrapped ice paks if the weather is particularly warm

Forms & Records:
http://www-heb.pac.dfo-mpo.gc.ca/intro_trans/form_b_e.pdf

References:
Egg Disinfection
Gamete Collection (Egg Take and Milt Collection)
Equipment disinfection
Federal-Provincial Introductions and Transfers Committee web page

Incubation

A basic understanding of egg development can be of great use in understanding the incubation requirements of those eggs. Salmon and trout eggs become progressively more fragile during a period from roughly 48 hours after water hardening until they reached the eyed stage. The eggs should not be handled during this extremely sensitive life stage.

Once the eggs reach the eyed stage, they are quite resilient and can withstand handling. This is the point at which egg shocking and egg picking generally should take place. Regardless of their less delicate nature at this stage, eggs should still be treated with care to avoid undue stress or damage.

Eggs are a delicate life stage and there are a number of factors that affect their health and development. Light, temperature, and oxygen are the three primary considerations in incubation. In nature, salmonid eggs are buried safely in redds, in cool, flowing, oxygen rich waters. In culture, we must attempt to mimic these conditions as best we are able to ensure high quality fry and good survival rates. In nature, the water in which eggs rear is exposed to many different pathogens and mortality rates to hatch are often high. In culture, we can protect the eggs during incubation from this early mortality through simple protective methods and appropriate disinfection procedures to prevent the introduction and/or spread of disease.

Predicted embryonic development times for five species of Pacific salmon and steelhead trout, using the models listed in Table 3 from Billard and Jensen (1996). Taken from Clarke 1997.

Species	Temperature ° C	Yolk plug closure		Eyed stage		50% hatch	
		Days	ATUs (°C-days)	Days	ATUs (°C-days)	Days	ATUs (°C-days)
Chinook	5.0	26.7	133.5	51.5	257.5	102.4	511.8
(O. tshawytscha)	7.5	17.9	134.5	34.2	256.6	70.3	527.5
	10.0	13.4	133.5	24.9	249.2	52.6	526.4
	12.5	10.6	132.1	19.2	240.5	42.1	525.7
Chum	5.0	31.9	159.6	50.1	250.3	99.6	498.2
(O. keta)	7.5	19.3	145.1	32.4	243.3	72.3	542.3
	10.0	13.3	133.0	22.9	229.0	54.4	544.5
	12.5	9.9	123.2	17.1	214.1	42.7	533.2
Coho	5.0	22.8	114.1	46.1	230.6	93.6	467.8
(O. kisutch)	7.5	16.3	122.1	31.5	236.6	63.1	473.6
	10.0	12.0	119.7	22.8	227.8	45.9	459.5
	12.5	9.0	112.9	17.1	214.4	35.6	444.8
Pink	5.0	36.7	183.4	51.4	257.2	109.0	545.0
(O. gorbuscha)	7.5	22.2	166.2	32.3	242.5	80.9	606.4
	10.0	15.1	151.5	23.1	231.4	63.0	629.6
	12.5	11.2	139.4	17.8	222.7	54.0	674.9
Sockeye	5.0	27.3	136.4	48.2	240.9	122.8	613.8
(O. nerka)	7.5	18.3	137.0	34.3	257.2	90.5	679.0
	10.0	12.6	126.0	25.0	249.6	69.3	693.2
	12.5	8.9	111.4	18.5	231.7	55.4	692.5
Steelhead	5.0	17.6	88.0	34.3	171.4	70.7	353.4
(O. mykiss)	7.5	11.7	87.5	23.9	179.5	47.2	354.0
	10.0	8.5	84.6	17.1	171.0	32.9	328.6
	12.5	6.5	81.1	12.5	155.9	24.8	309.8

Billard, R., and J.O.T. Jensen. 1996. Gamete removal, fertilization and incubation. Pages 291- 363 *In:* W. Pennell and B.A. Barton, Editors. Developments in Aquaculture and Fisheries Science V. 29: Principles of Salmonid Culture. Elsevier, Amsterdam.

Clarke, C. 1997. Predictions for salmonid egg development. Aquaculture Update No. 80. Fisheries and Oceans Canada. http://www.pac.dfo-mpo.gc.ca/sci/aqua/AQ/aq80.pdf


Predictions for egg development.pdf

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IncubWin:Predicting Embryonic Stages

4.7 Fertilization & Incubation

Rationale: The micropyle is in its greatest open position at the time the egg is taken from the fish. This small appendage commences to move to one side as soon as water entering through the pores starts mixing with the perivitelline fluid and fills the void between the outer shell and the yolk membrane. As soon this occurs, the opening decreases in size and continues to do so until the micropyle has moved into a position which completely seals off the opening to the outer shell. Therefore the possibility of fertilizing the egg gets progressively poorer as the micropyle opening get smaller, and since the spermatozoon enters only through the micropyle, fertilization is impossible after sufficient time has elapsed to allow the micropyle to close. Therefore, dry fertilization is recommended to ensure the greatest degree of fertilization prior to the addition of water. The goal of this SOP is to ensure hygienic and effective fertilization of eggs.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Eggs should be dry fertilized to increase fertilization success and reduce the introduction of potential water borne pathogens into the egg. Where possible, pathogen-free water should be added to the egg/milt mixture to activate the sperm. In general, well water is considered to be the cleanest water source on most facilities.

Assuming an equal proportion of males to females, an equal number of males and females should be killed and spawnings should be on a 1:1 basis unless matrix spawning is planned.

Following fertilization and washing, eggs should be disinfected to reduce the possibility of external pathogens entering the incubation system.

Details of the Procedure:

	Chinook	Sarita CK	Chum	Coho
Spawn Ratio (F:M)	**1 : 1**	**1 : 1 *Matrix***	**5 : 2**	**1 : 2 *Matrix***

1. Set incubation container flow rates to appropriate level for each container type (see below)
2. Set up the incubation room equipment (see below)

4.7.1 *Setting Incubation Flows*

During early January 2004, flows through the heath trays were checked and found to have significant (10% - 20%) variation between head tanks, as well as, with the "pre drilled" down spout caps. Head tank alarm probe 'activation' (positioning) has also contributed to lower (than acceptable) flows through the incubators.

With Heath stack inflow down spouts at a slight angle and 1/2" - 3/4" spill at the outflow plate of Head tanks # 8 - #16, the following cap colors and range of flows have been determined.

Yellow cap	9 L/min - 11 L/min (first incubation flow)
Green cap	14 L/min - 16 L/min
Black cap	17 L/min - 18 L/min
No Cap	18 L/min - 21 L/min

Factors such as container size and reaction time account for the flow variation within a cap color. This variation is acceptable within our first and second incubation standard loading of 2.5kg/tray for Coho and 3.5kg/tray for Chinook.

During annual incubation room set up, flows to all incubators will be checked with follow up random checks done during the season.

Alarm probes will also be checked and set to a level just slightly below the water surface of each head tank to all incubators.

With the water height in Head Tanks 12 / 13 set to the mark at the inflow end, the green and yellow caps were tried to see what flow each would allow. It was discovered that there were three different hole sizes drilled in each cap. It was also discovered that the position of the down spout could make a difference of up to 16.5% reduced flow in the straight vertical position. This verified the need to place the down spout in an angular position. Adjust so that there is no spillage over the back of the tray. This position must allow no restriction of flow caused by the down spout being too close to the sides or bottom of the tray.

After many measurements of flow carried out by two people with stop watches, the following colours and matching hole sizes complies with the operation standards for Health trays at Nitinat Hatchery.

White	[no hole]	flow 'capped off.'
Yellow	[7/16” drill]	flow = 11 L/min
Green	[17/32” drill]	flow = 15 L/min
Black	[9/16” drill]	flow = 16.4 L/min
No Cap	[¾” pipe nipple]	flow = 21.8 L/min

Other issues to consider is the tightening or loosening of down spouts, over flow required and alarms. This is the responsibility of the person setting up the head tank and should be completed on start up:

- If the down spout is too loose it will fall in the vertical position which may cause a restriction in the flow
- If the down spout is too tight, twisting could cause the elbow to split or strip threads.
- Head tank height and over flow should be set with the gate at the outflow end of the head tank with no more then ½” as a maximum
- Alarms must be adjusted to come on with a loss of head no greater then 1”

INCUBATION FLOWS *(measured in Liters per minute*)

INCUBATION CONTAINER	1ST INC	2ND INC	
HEATH TRAYS	7.5 - 8.2	18	per stack
ATKIN CELLS	30	60	per line
INCUBATION BOX	90	220	per line
KEEPER BOX		200	per box
KEEPER CHANNEL		400	per channel
SHALLOW POND		*900*	per channel

Optimum INCUBATION LOAD RATES *(kg of eggs)*

INCUBATION	1ST INC		2ND INC		
CONTAINER	WELL	RIVER	WELL	RIVER	
HEATH TRAYS	3 kg or 1 female	3 kg or 1 female	3 - 3.5 kg	3 - 3.5 kg	per tray
ATKIN CELLS	50 kg	45 kg			per cell
INCUBATION BOX	310 kg	270 kg			per box
KEEPER BOX			67 kg		per box
KEEPER CHANNEL					per screen
SHALLOW POND					per slat

4.7.1.1 FLOWS: (All Flows For First Incubation Will Be Set With Well (ground water)

Heath trays: flows are initially set at 8 litres/min. To accomplish this, only the small spigot or cap is turned down. As the eggs near pre-hatch (Second Incubation), the flows should be set to 17 litres/min, by turning only the large spigot down, or by placing the large cap on the spigot. The head tank outflow level determines the actual flow through the spigot.

Modified Atkin Cells: Flows have been set to 30 litres/min/line with the water depth in the head tank at the blue level line. This is located at the stand pipe end of each Head Tank. Prior to start up, check all of the flows. If adjustments are required, take the valve handle and adjust accordingly. Ensure the level in the Head Tank is maintained.

Incubation Boxes (Bulk Boxes): Set the flows at 90 litres/min/line.

4.7.2 ***Incubation equipment Set up***

4.7.2.1 Egg Counting Sheet:

The aluminium egg counting sheet is to be next to the cell that will receive eggs. On this sheet will be the talley counter and taped glass rod for counting sub-samples.

4.7.2.2 Clip Boards:

Plastic clip boards with the data sheets for each cell or box, and a pencil, will be hung on each corresponding Head Tank through-out the incubation room.

4.7.3 ***Planting Pathway***

Sarita Chinook: are planted first (start late Sept.) and will start at Atkins cell 6-1-C (a and b are empty for shocking) and progressing to 7-2-A, 7-2-B, 7-2-C etc. The last cells to be used for Sarita will be 7-4-D.

Note that the first two cells for this species are left empty for shocking and picking into latter.
Note: the cells used for Sarita Chinook must be clearly marked to prevent any mix-ups.

Nitinat Chinook: are planted starting in early Oct. The starting point will be Box7-1C, Progressing to 7-2-A, 7-2-B, 7-2-C etc.

Note: that the first two cells for this species are left empty for shocking into and picking into latter. When head tank 7 is full or if extra space is needed because of small eggtakes move to Head Tank 6, (Modified Atkins Cells) starting at cell 6-5-A and progress to 6-8-D.

Nitinat Chum: are planted starting Oct. 20. The starting point will be Box 0-1-A, progressing to 0-1-B, 0-1-C etc. Note that boxes, 0-0-1 & 0-0-2 Big Q. style for this species are left empty (for shocking into and picking into latter). When Head Tank 0 is full carry on to Head Tank 1. Start at Box 1-1-A and proceed 1-1-B etc.

Note: there is no need to leave any empty Boxes when starting new Head Tanks as long as First Incubation is going to use Well Water only.

If it is decided to use River Water in any Head tank for First Incubation the first two cells for this species are left empty for shocking into and picking into latter. When head tank 3 is full or if extra space is needed because of small eggtakes move to Head Tank 4, (Modified Atkins Cells) starting at cell 4-1-C and Progress to 4-8-D.

Nitinat Coho: are planted starting in mid Nov. through to late Dec. These are planted into Heath trays until eyed, at which point they are to be shocked and picked into Atkins cells (Head Tank 6) after Chinook have been moved out of 6 and 7 (leaving space for second incubation pathway for Nitinat coho from the heath trays to the cells.

4.7.3.1 Loadings and Water Source

Water Source:

Sarita Chinook are all on Well Water (ie Head tanks 6).
Nitinat Chinook are all on Well Water (ie Head tanks 7,6).
Nitinat Chum are all on Well Water (ie Head Tank 0, 1, 2, 3, 4)
Nitinat Coho are all on Well Water (ie Head Tank 5)

First incubation Loadings: All loadings are by weight

The following loadings are approximate and the actual weight goes to the nearest whole pail of eggs. (Do not break up a pail of eggs between two containers).

Container Style	Well
Bulk Incubation box	310 kg/box
Modified Atkin cell	50 kg/cell
Heath trays	3kg (Chinook) or 1 female/tray (coho)

4.7.4 *Gamete Transport*

Eggs: are to be transported, unfertilized, to the Incubation room . Each group of eggs are to be identified as to: 1- from where they came; 2- species; 3- the number of females.

Note: Chinook eggs are packaged one female worth of eggs per kitchen catcher bag if being transported from the field.

There will be two or three bags of eggs per 11 liter pail tied securely with air entrapped in each bag. Pails are to be stacked neatly in the incubation room as directed by the Incubation person.

Chum: are to be transported, unfertilized, to the Incubation room. Each group of eggs is to be identified as to: 1- from where they came; 2- species; 3- the number of females.

Note: The eggs from five female chum are packaged per kitchen catcher bag when being transported from the field.

There will be two bags of eggs per 11 liter pail tied securely with air entrapped in each bag. Pails are to be stacked neatly in the incubation room as directed by the Incubation person.

Sperm (milt): is to be kept cool, dry and dark, and brought to the Incubation room. Each group of sperm is to be identified as to:1-from where they came;2- species; 3- # of males.

Note: Chum and Chinook sperm are packaged one male worth of sperm per whirl pack bag. Bags of sperm are to be in a pail or box cooler with burlap covered ice packs. Pails are to be stacked neatly in the incubation room as directed by the Incubation person.

4.7.5 *Fertilization and Planting*

Sperm may be activated by ovarian fluid and/or water. Both methods are utilized at Nitinat Hatchery with the water being the second stage. Using the ovarian process as the first stage allows one male to be mixed to one pail of eggs, giving this male an excellent chance of fertilizing these eggs. However, if this male is sterile or the sperm is dead, when the eggs are poured into the weighing pail they are mixed with other pails of eggs and milt. This will allow activated sperm from other males to have a chance of fertilizing eggs which may have been mixed with dead sperm. The overall result, is a better fertilization rate. This procedure should avoid the problem of sterile males, yet enable the Hatchery to meet allowing the male to female spawning ratios required, therefore maximizing genetic diversity.

Eggs:

1. Take a pail containing bags of eggs and place the pail onto the cart table.
2. Gently take a bag of eggs from the pail and place it into a dry pail.
3. Taking the scissors carefully cut off the corner of the bag (a good sized cut).
4. Lift the far corner of the bag so that the eggs pour out of the bag into the pail, (do not allow the eggs to drop into the pail) try to get all the eggs out without breaking them (take special care with eggs stuck in the bag's knot).
5. Only one bag per pail is allowed during this process (which means for Chinook one female per pail, and for chum five females per pail). This will permit ease of inventory control.
6. Repeat this process ensuring each new pail is clean and dry.

Sperm:

1. Take a Whirl-pak of sperm (one male) use the scissors to cut off the tied end.
2. Pour the sperm into the pail of eggs spreading the sperm over as many of the eggs as possible. (If the pak is full of sperm do not use more than 15 ml of the sperm. The rest can be discarded or, if so ordered, used for another pail of eggs).
3. Sperm is stirred into the eggs using one hand, with your other hand another pail can be stirred if required.
4. Gently and slowly mix the eggs and sperm for about 8 seconds. (It is highly recommend that surgical gloves be worn to prevent sperm burn).
5. Care must be taken that a thorough mix is obtained ensuring the eggs on the bottom of the pail have come into contact with the sperm.

NOTE: always use fresh sperm when possible. Do not use overnight sperm or that suspected of being contaminated, ie: bloody, green,etc.

Weighing the eggs:

1. The pails with eggs and sperm are now carried over to the weighing cart.
    a. In the case of Chinook, three pails of sperm coated eggs are poured into the pail.
    b. For Chum, two pails of sperm coated eggs are poured in the pail on the scale which has previously been tared (zeroed).
2. As each pail of eggs is poured into the pail on the scale, click the talley counter placed in front of the weigh scale. This will keep track of the number of pails and therefore the number of females and males used (make sure you click in every pail as you will never remember this simple number).
3. Record the displayed weight onto the data sheet; and use the counter number to record the number of females and males used onto the data sheet.

Sub-Sample Egg Weight:

1. Take the sub-sampler beaker and dip into the basin, this will initially coat the beaker with ovarian fluid and sperm, pour out any eggs and surplus fluid.
2. Place the coated beaker onto the smaller sub-sample weigh scale and tare (zero) the scale.
3. When weighing each pail of mixed eggs and sperm, take out between 5 and 10 sperm coated eggs and place them into the prepared sampling beaker.
4. When the box or in the case of cells when a row is complete, record the displayed weight of eggs onto data sheet (Record it. You will never remember this weight).
5. Next take the beaker of eggs over to the receiving box or cell where there should be the Egg Counting Sheet, a glass rod and a tally counter to count the eggs into the receiving cell. (remember to record the number of eggs in the sample and the location number of the cell).

Pouring Pails of Eggs into Boxes or Cells:

Once pails have been weighed and the data has been recorded onto the data sheet, carefully remove the pail of sperm coated eggs from the scale and pass it to the egg washing crew.

Manual Egg Washing:

<u>The egg washing crew must complete the following steps in order:</u>

1. Gently add water to the pail from the overflow of the last box or cell in a row.
2. Give the eggs a gentle stir by hand
3. Leave the eggs in this water for a minimum of 15 seconds
4. After 15 seconds pour off the water being careful not to lose any eggs
5. Refill the pail with water
6. Stir the eggs
7. Pour off the water and contaminants (ie: blood clots, broken egg shells, pieces of fish flesh, pieces of skein and cloudy water)
8. Repeat washing until there are no contaminants left.
9. Pour the pail of eggs into their proper location using a swirling motion.

Note: It is extremely important to wash and plant the eggs properly, as no other form of fungus control will be used during incubation!

10. Once the box or cell is full, gently level out the eggs.
11. Remove any sperm, blood, etc. which collects on the outflow screen.
12. When the pail is empty, place it under the out-flow of boxes or cells to rinse.
13. Repeat the above steps using a different data sheet for each cell or box.

Labour:

The person in charge of first incubation is responsible for ensuring that all the above steps are being followed. These steps, under most situations, can be performed by three persons, except for very heavy egg take days, when more bodies will be required.

The general division of labour is to have one assistant open the bags, add sperm, mix, & pass the pail of eggs to the weigh station. The person in charge of first incubation is to do the most important, disciplined steps of weighing eggs, weighing sub-samples and recording data.

The washing and pouring of the eggs into the boxes or cells is to be accomplished by washing crew.

The counting of the sub-sample, and recording of this data, is to be carried out by one designated person only. This will allow the person in charge time to organize their thoughts & monitor the whole process, and direct shipments of eggs coming into the incubation room.

Lulls:

When all the pails have been processed, and there will be no eggs for a period, perform the following tasks:

With 5 minutes of free time:

- Clean equipment
- Hose the floors down
- Arrange cell covers

With 15 minutes of free time, when jobs above are complete:

- Have crew go to ponds and help egg take crew.
- Do marked recovery and counts from fish totes.

- The person in charge of first incubation is to:
  - Go to the office and have the collected data entered onto the computer.
  - Time permitting, assist the person in charge of egg takes.

4.7.6 *Confirmation of Inventory*

Always during, or at the end of the day, confirm your adult numbers and where they were from with those of egg take leaders at the hatchery or in the field. (The usual problem is the number of females and males used, so negotiate and adjust numbers reasonably). Two complete weighing carts with scales are available, so one may be set up for Chinook and the other set-up for chum. This is especially helpful when the two species overlap in timing.

4.7.7 *Incubation*

DO NOT DISTURB THE EGGS UNTIL THE EYED STAGE. At roughly 25 ATUs an egg dropped 4 mm, has a 50% chance of dying.

When the eggs near 200 ATUs check on a daily basis to see when the very first eye can be recognized, and record this date as the date and ATU at First Eyed (normal appears to be around 220 ATU).

4.7.8 *MATRIX SPAWNING*

To ensure genetic diversity within a species containing low numbers of spawning adults, a system of splitting up milt or eggs must be used. At Nitinat Hatchery matrix spawning is used when fertilizing Sarita Chinook and Nitinat Coho. Hatchery staff will endeavor to capture at least two males for each female. When fertilizing the female's eggs, they are to be split into two equal parts. Each part is to be fertilized with a different male. After a male's milt has been used, any remaining milt from that male is to be discarded. If by some chance there is more than a 2:1 ratio of males to females, split the female's eggs up into equal lots totaling the number of ripe males.

EXAMPLE: Matrix Spawning using 4 Males and 4 Females:

	M 1	**M 2**	**M 3**	**M 4**
F 1	F1 x M1	F1 x M2	F1 x M3	F1 x M4
F 2	F2 x M1	F2 x M2	F2 x M3	F2 x M4
F 3	F3 x M1	F3 x M2	F3 x M3	F4 x M4
F 4	F4 x M1	F4 x M2	F4 x M3	F4 X M4

4.7.9 *Sodium Bicarbonate rinse*

Sodium bicarbonate is used as in solution as an egg rinse to aid in fertilization

Equipment required:

1 large garbage bucket
Weigh scale
Stir stick

Mixing procedure:

1. Weigh out Sodium Bicarbonate at 13 g/L of water.

   i.e. 100 liters of water requires 1300 grams of Sodium Bicarbonate

2. Place Sodium bicarbonate into garbage bucket
3. Add appropriate amount of water (100 liters)
4. Mix well

Procedures for use as an aid to fertilisation

1. Weigh eggs
2. Add sperm to eggs
3. Mix eggs and sperm gently
4. Add bicarbonate solution to eggs
5. Mix gently
6. Wait 90 seconds
7. Wash eggs
8. Plant eggs
9. Discard all unused solution at end of day

To minimise water hardening of eggs before fertilisation

- Rinse egg buckets in solution before use.
- Rinse hands with solution before handling eggs.

4.7.10 ***Chiller Setup and Shut Down***

Chiller Set up

1. *Pre read background information on "New Ocean" titanium chillers/chiller-heaters*
2. Start and fill ambient head tank (HT) #3 with ground water and adjust supply valve to 100 L/min. (A1)
3. Set flow on "make up" supply valve to 10 L/min.(A3)
4. Adjust level in HT#3 to ¼" above standpipe and S*et Alarm*
5. Swing ambient supply to upright (off) position (A2)
6. Install "grey" drain plug from under side of HT#4 and turn on ground water supply, filling Head Tank #4, Incubation and Recovery boxes, once full turn off.
7. Turn on pump at breaker (*In panel "HA" pump will start automatically*)
8. Balance HT#4 supply *(C* 3 *& C4*) to "just" overflowing to maintain head.
9. *Set Alarm*
10. Connect cooling water supply to Chiller cooling inlet. Safety wire in the "on" position.
11. Turn on chiller at breakers in Panel *"HA"*

Chiller Shut Down

1. Turn off Chiller at control panel
2. Turn off valves A1 & C1 on HT# 3 & 4 at Incubator inlet.
3. Turn off Chiller and Circulation pump breakers in Panel “HA” and lock out. (Note pump will start automatically when power is restored)
4. Turn off make up water supply line. (*A4*)
5. Turn off and disconnect cooling water supply
6. Drain and clean boxes & pump.
7. Place all associated troughs and plumbing accessories into recovery box.

Chiller Operation

1. Turn on chiller and set desired temperature.
2. Ensure *Temprinter Probe* (P3) is installed and operating on *max/min and averaging* mode.
3. Monitor over flow from recovery box. Chiller will cool to temperature setting in approximately 6 hours. (Note there should always be overflow from recovery box)

4.7.11 ***Nitinat Chinook and Chum***

Nitinat Chinook and chum are spawned off the sorting table into buckets. The sperm is added at the egg take shelter and egg washing occurs once the fertilized eggs have been moved into the incubation building.

Fertilized eggs are generally planted into bulk boxes unless there are not enough eggs to fill one over a 3 day period. (Between 500,000 to 750,000 eggs are planted into each bulk container). Where there are insufficient eggs to fill a bulk incubator to within 2 inches of the lip of the container, eggs are planted to an Atkins cell.

4.7.12 ***Sarita Chinook***

The Sarita Chinook egg take is smaller, involving less fish.

1. Set up appropriate Atkins cell and fill with egg disinfecting solution (Ovadine – 100ppm)
2. Eggs and milt are taken separately and eggs are fertilized after moving the gametes into the incubation room
3. The bags of eggs are opened and the eggs are gently spilled into buckets (fertilize batches of 25 – 30 females at a time)
4. Two staff members are responsible for weighing and subsampling eggs to determine the inventory
5. After subsampling, the Whirl-paks of milt are opened and the milt from one male is deposited into the appropriate number of buckets of eggs as determined by matrix spawning plan

6. Mix gently by hand
7. Add water to each container of eggs and milt once the series of containers has been fertilized
8. Decant the water into a tote and repeat washing as needed to remove contaminants (blood, faeces)
9. Gently pour the fertilized, washed eggs into the appropriate Atkins cell (Head tank 6)
10. After the eggs have been in the Ovadine solution for 10 minutes, turn on the flow to the incubator to flush the solution out

4.7.13 *Coho*

Coho are treated as above with the exception that eggs are immediately planted to Heath trays (how many females per tray…onne??)

When the diagnostic results are returned by the pathology lab, eggs from any female that tested as a low positive or a negative will be inventoried from Heath tray into an Atkins cell. Low positives are used for outplants, only negatives go into the production program

Any eggs that are identified as originating from a moderate or a high positive female are discarded. Discarded eggs are frozen for disposal to a composting facility in Port Alberni.

4.7.14 *Second Inventory*

When the second incubation containers are to be loaded, eggs are dipped from the appropiate box/cell and weighed on the large scale. This weight is recorded, along with the date and where the eggs were moved to.

The weights achieve two purposes:

1. The first is the weight of eggs in the cell, this can then be used along with the live and dead egg enumeration , to calculate the number of eggs in the cell, and back calculate the "First Real Inventory"
2. The second is the weight of eggs going to the next incubator (ie; keepers or boxes. This weight is different from that of each box/cell because a box/cell may be split and some eggs go to one incubator and some eggs to another incubator. This weight is used to calculate the number of eggs going eventually into a pond.

4.7.15 *Transfer of Eggs to Second Incubation*

When the eggs have been weighed:

Sarita and Nitinat Chinook and Nitinat coho are inventoried into the Health Tray baskets

Nitinat chum:

1. Weigh the eggs into baby basins.
2. After weighing, place the basins on the 6-wheel carts.
3. Take the carts to the new incubator (keeper box or channel) and leave them for egg planters to spread the eggs.
    a. Egg planters will gently spread the eggs on the screens starting at the bottom of the channel
4. While one cart is in use, another cart is being loaded with basins to keep a good flow of eggs to the egg planters.
5. Generally each basin will have about 7kg of eyed eggs. These weights may vary due to planting and keeper loadings required to meet operational objectives.

Forms and Records:


Flow Rates July 2001.xls


Incubation.xls


Coho Incubation.xls


2nd Incubation Log.doc

References:

Egg Disinfection


Genetic Practices


Planting Plan.XLS

4.8 Egg Disinfection

Rationale: Eggs can be safely disinfected following fertilization, during or after water hardening. The purpose of egg disinfection is to minimize the pathogen load which may have come from the broodstock and decrease subsequent spread of pathogens between eggs or egg batches. This SOP addresses section *2.1.3.7* of the General Principles of Fish Health Management. The goal of this SOP is to ensure safe disinfection of eggs following fertilization.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Ovadine™ is commonly used at fish hatcheries for equipment disinfection. It has also been safely used for over two decades as an egg surface disinfectant during water hardening.

Ovadine™ is currently (2007) under review by the Veterinary Drug Directorate for approval as a fish egg disinfectant. Until approval is received, it is available only by prescription from a licensed Veterinarian, through Health Canada's Emergency Drug Release (EDR) program (see Appendix for Ovadine™ Emergency Drug Release (EDR) – Hatchery Reporting Requirements information sheet)

Eggs may be disinfected at water hardening after fertilization, after egg picking, and after eyed eggs are transferred to a new site.

Disinfection should not been done within 5 days of hatch, as it can stimulate premature hatching with increased mortalities.

Eggs are treated with a 100 ppm iodine solution for 10-15 minutes. A 100 ppm concentration of Ovadine™ is made by adding 10 mls of Ovadine™ to each litre of water.

The actual volume of each incubator should be predetermined before starting.

A suitable ratio is 1 volume of eggs to 10 volumes of disinfectant solution.

Ovadine solutions will be a rusty brown colour when fresh, but as the iodine degrades, the solution will start to lighten in colour to yellow indicating a loss of activity and effectiveness. If the colour lightens before time has expired, additional Ovadine can be added and mixed gently to ensure even distribution.

Local waste management regulations regarding safe disposal should be followed. Diluting spent Ovadine™ bath solutions with the rest of the effluent from a facility is considered sufficient before discharging to a stream; however, if dilution is not possible, it can be safely disposed to ground.

Details of the Operating Procedure:

Sarita River Chinook and all coho eggs are disinfected.

Equipment

"Ovadine" (Povidone Iodine – 10% - 12% Active Ingredient) solution purchased from Syndel International or Dynamic Aqua in 20 liter quantities.

Large (100L/+) Garbage pail on wheels

Stopwatch

Large Graduated beaker or flask

Stir stick

Procedure

1. Read MSDS sheet (under "O") in binder in lab and take appropriate H&S measures when handling product.
2. Mix enough solution for the day or mix 1 liter (1000ml) of Ovadine with 100 liters of water and stir well.
3. Remove lids and water from trays and add 5 liters solution to each tray. When using Atkin's cells approximately 50 liters of solution will be required for a full cell of eggs.
4. Wash eggs after fertilisation and place 1 female (coho) in each tray or a maximum of 45 kgs. in each Atkin's cell.
5. Ensure eggs are covered with solution and lids are in place on heath trays
6. Eggs must remain in Ovadine for a minimum of 10 mins. Timing for Atkin's cells begins when the last female is added. For a full cell (20-25 females) several egg washers are required.
7. Carefully push trays in after 10 mins. Atkin's cell above "treatment cell" should be full of water when flow is restored.
8. Dilute remaining Ovadine and discard to drain.

Forms & Records:
Error! Reference source not found.

References:
Ovadine™ data sheet: http://www.syndel.com/d_p_f_s/ovadine_info_sheet.html
Veterinary chemicals: http://www.dfo-mpo.gc.ca/science/aquaculture/aah/veterinary_chemicals_e.htm

4.9 Egg Fungal Treatments

Rationale: Dead eggs serve as growth media for saprophytic fungal infections. Once a fungal infection has started, it rapidly spreads to adjacent eggs and can result in poor survival to hatch. Egg disinfection and picking (see *Egg Disinfection* and *Egg Shocking, Picking & Egg Enumeration*) are the first steps in preventing fungal infections. However, depending on water source, temperature and hardness, preventing and controlling fungal infections of eggs may be best accomplished by administering chemotherapeutants. The goal of this SOP is to safely manage fungal infections of eggs.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

The fungal treatment program is determined by the Fish Health Management Team. Management and will be site-specific according to treatment capabilities, historical egg fungus infection rates and egg survival data.

Staff administering chemical treatments to eggs is aware of WHMIS safety information and employs appropriate personal protective equipment.

Egg batches should be observed on a routine and frequent basis to assess and track the development of mortalities and fungal infection.

Approved chemotherapeutants for egg disinfection include: Parasite-S™ a.k.a. formalin (a liquid solution containing 37% formaldehyde), Perox-Aid™ (a 35% hydrogen peroxide solution). Formalin is generally the preferred treatment.

Parasite-S ™ a.k.a. formalin

Formalin treatments can vary in concentration from 1000 to 2000 ppm. The duration of exposure is generally 15 minutes, applied as a constant flow to incubation water supply. The scheduling can be repeated as needed to control fungal infections. **The standard treatment for eggs is 1670 ppm formalin for 15 minutes twice weekly.** Formalin should be metered in with a reliable delivery system.

Treatments can be started a day or two after fertilization and may be continued throughout incubation; however treatments should be stopped at least 5 days before anticipated hatch to reduce the risk of stress-induced premature hatching. Alternatively, treatments can be discontinued after shocking, if egg picking alone can prevent fungal infections from developing.

When calculating the treatment dose of Parasite-S, the product is considered 100% active. 1670 ppm (or 1.67 ml/L) is the equivalent of a 1:600 dilution. This can be calculated as:

1000 ml//600 = 1.67ml/L

Formalin effluent restrictions: Depending on temperature and species, concentrations of formalin > 200 ppm can be toxic to fish. Effluent target concentration is < 25 ppm formalin; this dilution should be achieved by combining incubation flows with the discharge from other rearing units prior to release into a natural watercourse.

Staff should review WHMIS information prior to handling this product and employ appropriate personal protective equipment. Formalin must be handled with care. It is harmful if inhaled, and can seriously irritate eyes and skin after contact. Formalin should only be handled in well ventilated areas, preferably while wearing a respirator and safety glasses.

Perox-Aid ™

Hydrogen peroxide treatments can vary from 250 – 500 ppm. **The standard treatment regime to prevent fungal infections of eggs is 500 ppm for 15 minutes every other day.** To treat existing fungal infections, 500 ppm for 60 minutes every other day may be used. However, existing fungal infections will be better controlled with Parasite-S.

PeroxAid volume calculations are based on a 35% active ingredient (hydrogen peroxide concentration). So to obtain a final concentration of 500 ppm:

$$\text{Volume of PeroxAid per litre} = \frac{\text{recommended dose}}{\text{active ingredient}}$$

$$= \frac{500 \text{ ml/L}}{35\ \% \text{ or } 100/35}$$

$$= 143 \text{ ml PeroxAid per litre}$$

In its concentrated form, Perox-Aid is a strong oxidizer that can burn skin or membranes. Staff should review WHMIS information and use appropriate personal protective equipment when handling this product.

Pyceze ™ (Bronopol)

Pyceze™ is an antimicrobial agent only available through veterinary prescription and Health Canada's Emergency Drug Release Program for antifungal treatment of salmonid eggs.

Salt

Sodium chloride has historically been used for egg fungal control, however this use is not approved by Health Canada.

Details of the Operating Procedure:

Treatments are carried out on Mondays and Thursdays

4.9.1 ***Bulk Box Head Tanks***

1. Treatment concentration is **1700 ppm**
2. Adjust flow in head tank so that there is NO overflow
3. Measure 3 liters of Parasite-S to treat a flow of 90 liters/min through a bulk box line. Each 3 L volume will treat one line.
4. Add the 3L of Parasite-S to the metering pail and dilute with water to the 20 liter mark in the metering pail.
5. Turn on the pump and ensure that the solution is being dispensed into the line
6. Dispense the contents of the 20 liter pail using the Manostat pump over 20 minutes.

***Note:** **Use caution with this treatment and wear hand and eye protection. Read the MSDS sheet for this product prior to using.**

4.9.2 ***Atkin Cell Head Tanks***

1. Treatment concentration is **1700 ppm**
2. Adjust flow in head tank so that there is NO overflow
3. Measure 1 L of Parasite-S to treat a flow of 30 L/min through a bulk box line. Each 1 L volume will treat one Atkins cell line.
4. Add the 1L of Parasite-S to the metering pail and dilute with water to the 20 liter mark in the metering pail.
5. Turn on the pump and ensure that the solution is being dispensed into the line
6. Dispense the contents of the 20 liter pail using the Manostat pump over 20 minutes.

***Note:** **Use caution with this treatment and wear hand and eye protection. Read the MSDS sheet for this product prior to using.**

Forms & Records:

Parasite S Treatment Form.doc

References:

Parasiticides and Fungicides: http://www.syndel.com/d_p_f_s/parasiticides_fungicides.html
Perox-aid™ data sheet: http://www.syndel.com/d_p_f_s/perox-aid_info_sheet.html
Parasite-S™ data sheet: http://www.syndel.com/d_p_f_s/parasite-s_info_sheet.html

4.10 Egg Shocking, Picking & Egg Enumeration

Rationale: Dead eggs are removed to reduce fungal growth and disease transfer. This SOP addresses section *2.3.2* of the General Principles of Fish Health Management. The goal of this SOP is to ensure effective removal of dead eggs, which can serve as growth media for saprophytic fungal infections.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Egg batches are observed on a routine and frequent basis for mortalities. After eggs have reached the eyed stage, they can be physically shocked to allow the discrimination between viable and unviable eggs. Shocking will rupture the yolk (vitelline) membrane of eggs which are undeveloped or infertile and result in an influx of water turning the egg white. Dead eggs may be removed, or picked, as required to keep the proportion of dead eggs in the incubators to a low level.

Pre-eyed picks are only attempted if mortalities are very high. High mortalities are defined as when a great number of dead (white) eggs are visually observed during the fertilization process. Extra care must be taken to avoid disturbing live eggs while picking out dead eggs.

Eggs may be picked by hand using modified tweezers. Mechanical egg pickers are operated according to the manufacturer's specifications.

Regardless of the method, tweezers or mechanical pickers should be sterilized between egg batches (see *Equipment disinfection*).

Details of the Operating Procedure:

4.10.1 ***Egg Shocking:***

When a strong eye has developed, about a five days after eyeing, the eggs can be shocked. (Only the person in charge of first incubation or the person they appoint will be allowed to shock eggs).

1. To shock the eggs, use the long handled dip net to dip the eggs out of the box/cell.
2. Lift the eggs three feet into the air and gently, in a singular motion, pour the eggs into the empty cell forward of the cell they were removed from. (When shocking, ensure a motion is used which will cover the entire area of the box/cell).
3. Record the date the eggs were shocked and the location where the eggs were shocked to on the data sheet.
4. After shocking, check the eggs to see if there has been any disruption of the membrane (look for white eggs). This is an indication that the eggs are being shocking overly hard.

5. If this inspection shows eggs that have not turned white, yet look blank, shocking may not be sufficient.
6. Let the eggs sit for two full days before picking, this will allow time for the eggs to fully turn white, making picking more efficient.

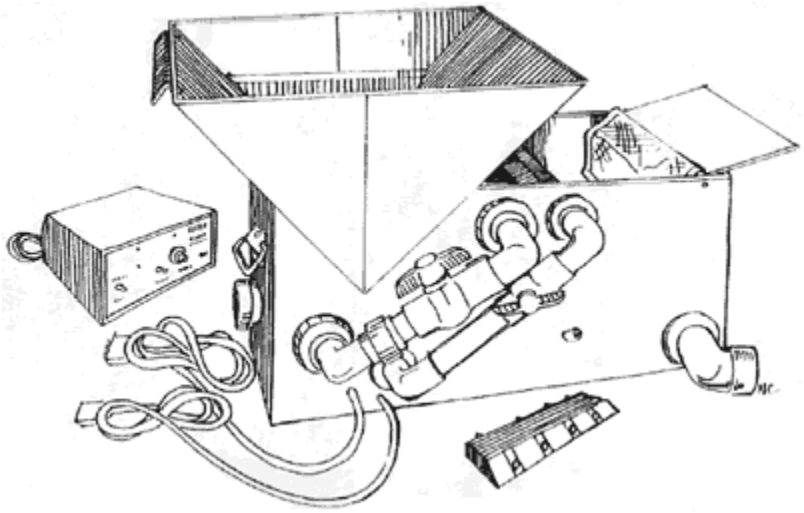
4.10.2 ***Egg Picking***

4.10.2.1 Hand Picking:

At times, some hand picking may be required. Spring tongs and suction bulb pickers are available from the fish culturists. Both require working with your hands in cold water. This process should only be done if the machines break down or if shocking of the eggs has been insufficient.

4.10.2.2 Machine Picking:

Eggs are picked using an 8 channel JM-8 Jensorter high speed egg picker.



Make sure all electrical and water connections are properly connected and the overflow dams are in place (see fish culturists).

Set up and calibrate the picker according to the manufacturer's manual of instructions.

4.10.2.3 Picking Process:

1. Dip the previously shocked eggs out of the cell and fill the hoppers of each machine.
2. When the eggs are picked, return the eggs to the forward cell; ie if eggs were taken from box 0-1-C they would be relocated to box 0-1-B.
3. Record the date and where these eggs have moved to.
4. With the machines working properly should be able to do about two boxes or 14 cells per day.

5. If the survival rate is lower than 87%, the machines will bog down, making many mistakes, and slowing the whole process down. In this situation, use one machine to re-pick the dead eggs from the other machines.
6. Also, every other hole on the egg picking disk may be taped over, this effectively cuts the speed in half but allows the machine to keep up with all the dead eggs, thereby saving time.

Labour:

1. Usually three staff are required to operate the picking machines and record data
2. One staff member loads the machines, another returns the eggs to the cell and weighs and sub-samples the dead eggs
3. A third staff member can be used for second picking, keeping the machines running properly and maintaining the area

4.10.3 *Dead Egg Enumeration*

1. Weight the container of dead eggs in a tared container
2. Record the weight on the data sheet
3. Remove 100 ml of dead eggs in a small glass beaker
4. Weigh the 100 m egg subsample
5. Record this data
6. Pour the eggs out onto a smooth surface and count the number of eggs in the 100 ml sample
7. Repeat this
8. Calculate the number of dead eggs from the each box or cell

4.10.4 *Live Egg Enumeration*

1. Dip a 50 hole egg paddle into the incubator to be enumerated. Ensure that all of the holes are filled and fill the holes, thereby automatically counting 50 eggs at a time
2. Pour the 50 eggs into a small container that has been tared on the scale
3. After each dip stir up the box/cell
4. Remove a total of 6 paddles worth of eggs (300 eggs), and place them into the tared container on the small scale
5. Record the weight of the 300 eggs on the data sheet
6. Repeat this process two more times (ie 3 sets 900 in total per box/cell)

Forms & Records:

References:

Equipment disinfection


Shock sensitivity in eggs.pdf

Clarke, C. 1997. Mechanical Shock sensitivity in salmonid eggs. Aquaculture Update No. 78. Fisheries and Oceans Canada. http://www.pac.dfo-mpo.gc.ca/sci/aqua/AQ/aq77.pdf

Jensen, J.O.T. and D.F. Alderdice. 1983. Changes in mechanical shock sensitivity of coho salmon (*Oncorhynchus kisutch*) eggs during incubation. Aquaculture. 32: 303-312.

Jensen, J.O.T. and Alderdice, D.F., 1989. Comparison of mechanical shock sensitivity of eggs of five Pacific salmon (*Oncorhynchus*) species and steelhead trout (*Salmo gairdneri*). *Aquaculture*, 78: 163-181.

4.11 Ponding

Rationale: Removing fry from incubators when 80-90% of fry have utilized 80-90% of their yolk-sac promotes growth and reduces fish health risks from early ponding. Not removing fry from incubators at this stage of development or not ponding fry based on a maximum wet weight measurement poses a risks to initiating proper feeding and to fish health from early ponding. The goal of this SOP is to ensure that fry are ponded at the appropriate time and in the appropriate manner to ensure maximum survival and transition to feed.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

Details of the Operating Procedure:

4.11.1 ***Ponding from Heath trays to asphalt and concrete ponds, small containers and circular tubs***

1. Check the ATU's on the fry and visually inspect the fry's development for ponding readiness
2. Set up the pond and adjust the flows as required [see supervisor]
3. Connect a 10 ft. section of hose from the supply valve [located above the ponding basin] placing the other end in the mixing section of the basin.
4. Supply enough water to completely fill the hose with a depth in the basin of no less than 18"
5. At the pond secure the hose leaving a drop from the hose to the water in the pond of approximately 6" to 18".
6. Check the ponding hose for leaks and trapped air bubbles (if either is found fix the problem before continuing)
7. Set up the tray cleaning stands
8. Set up the diesel pressure washer [see STEPS]
9. Using the 6 wheeled cart go to the last stack of Health trays to be ponded for the day .and work toward first stack of trays for the day
10. Remove trays in the stack containing fry and placing them on the cart and take them to the ponding location [see supervisor as to the number to be ponded at one time]
11. With the basket in the ponding container remove the lid and pour the fry into the water
12. All dead eggs or alevins remaining in the basket or on the lid must be collected for sampling at the end of the day when the results are recorded
13. While ponding is taking place, clean the trays, baskets and lids using the pressure washer.
14. Set damaged units aside for repair. Units in good condition can be returned to their stacks.
15. After a complete stack has been pond, inspect the fry in the pond to ensure everything is working properly. [after the first inspection it is only necessary to check every fourth stack]

16. Remember it is important to work slowly and only as fast as the slowest step. In most cases this is determined by *"swim up"* of the newly ponded fry
17. Leave the water running 24hrs before and flush the line before changing to another pond.
18. As a "rule of thumb" pond to a maximum of 500,000 fry/day

4.11.1.1 Heath trays to small containers

1. Set up the small containers with baffles screens, stand pipes and covers

4.11.1.2 Heath trays to concrete ponds

1. Attach the 90^{o} CAM-LOCK adapted elbow to the ponding basin and the ponding hose

4.11.1.3 Heath trays to asphalt ponds

1. Attach the 90^{o} elbow (with the garden hose valve attached) to the distribution [cam-lock] connection at the inflow end of the pond
2. Connect the ponding basin to the underground cam-lock connection located at the North East end of building # 3
3. Remove any trapped air in the underground distribution pipe by opening the garden hose valve

4.11.2 ***Ponding from Keeper channels and Keeper boxes (to concrete rearing ponds)***

1. Check fry for readiness by glassing in a beaker samples from various areas of the keeper and referring to the ATU sheet. Fry swim-up in the water column is also a good indication of readiness. Always consult with fish culture personnel prior to initiation of ponding.
2. Keepers to be ponded must have been flooded prior to proceeding with following steps
3. At concrete pond inflow, remove stop log panel
4. Place two 4" stop logs and "V" weir in outlet guides
5. Bolt "Y" trough or fry collection box in place
6. Cover "Y" trough with two egg screens (predators)
7. Ensure that "fry baffles" at the inflow are at their highest setting
8. Once fry readiness is confirmed, remove the bricks holding down the slats and stack neatly on the centre walls and along the outside keeper wall
9. Using the 6 wheeled cart and frame remove the slats to outside of the keeper building so they are ready for cleaning
10. Set up the pond with V screens, stop-logs, V notches and adjust the flows as required for ponding [see supervisor]
11. Count and record the morts before using a wash down hose to spray clean both sides of the slats and stack them on pallets along side of the building

12. Remove and store the 2” x 2” stop logs from the keepers to be ponded
13. Remove one 2’ x 4’ stop log from the outflow end of the keeper and place it in the stop log guides in the middle of the channel

Note: this will allow you to pull 1 stop log at a time and no spillage over the “ Y “ will take place

14. Pick the majority of the dead eggs and fry from the fry screens
15. Sample and record the morts
16. Remove the fry screen allowing fry to migrate to the pond
17. Over a 2 to 3 day period flush the fry to the pond by removing and replacing the 2 stop logs in the keeper

Note : never have both stop logs out at the same time until the very last flush and never leave the stop logs out for extended periods

18. When all fry in the keeper channel have migrated into the pond remove the “ Y “ trough or “collection” box.
19. Replace the pond inflow stop logs using foam and wedges to ensure they are fry tight
20. Adjust the pond to the operating height and flow [see supervisor]
21. Count the dead remaining in the keeper and record the number
22. Using water, clear the debris in the keeper channel out to the distribution channel
23. Turn off the water and let the keeper channel dry

Forms & Records:


Biomass.xlsx


container volumes and loading rates.xls

References:

Clarke, C. 1997. Predictions for salmonid egg development. Aquaculture Update no. 80. http://www.pac.dfo-mpo.gc.ca/sci/aqua/AQ/aq80.pdf

Rearing

Rearing constitutes the period immediately following ponding when feed is first offered until fish are released. This is obviously an important period, particularly in light of the fact that this is the period when the single most costly factor arises in hatchery production of fish, namely feed. If fish are not maintained in the healthiest manner possible, feed is being wasted as the fish partition energy into process other than growth.
Proper nutrition aids in growth and health, addition of immunomodulators may give an important boost to fish, especially during periods of stress such as handling, marking, higher than average water temperatures etc. The best foods available may lose their value through improper storage, therefore it is important that feed is maintained in a pest free, cool environment to ensure that fats and oils do not go rancid and that vitamins remain biologically available to fish. There is little point in spending money on fish feed if it is not cared for in a manner that ensures it meets the nutritional requirements of the fish.

Stress is a major factor in fish health and factors that result in stress to fish should be mitigated to reduce incidence of disease on any facility. Stress can result from many factors. Inadequate water quality accounts for more disease outbreaks than any other factor. High water temperatures, low dissolved oxygen levels, excessive suspended solids, nitrogenous waste build-up and a host of other factors can result in physiological stress which can funnel energy reserves away from the immune system reducing disease resistance in favour of maintenance of homeostasis. Improper protection of fish through the use of covers, predator netting or other deterrents may result in losses to predation and associated stress in surviving fish. Inadequate cleaning of enclosures can lead to biofilm development and may provide harbour for potential pathogens. These represent a fraction of the potential stressors that can occur on a fish culture facility and merely serve to highlight the importance of reducing stress during rearing.

While it is preferable to handle fish as little as possible, some handling is required to ensure appropriate daily rations and to avoid waste and associated reduction in water quality. To facilitate this, regular determination of average fish weight in an enclosure is necessary. It is important that representative samples are taken, thus it is important to take samples in a random manner with fish crowded to resolve any bias in the process.
In most cases, hatchery fish are marked as such through the use of fin clips and, in some facilities, the use of coded wire tags. Again, this is another handling procedure and in this case anaesthesia will be required to reduce the degree of stress on the fish during the process. As in all handling procedures, care should be taken to minimize stress and any possible damage to the mucus coat of the fish, both of which can lead to an increased susceptibility to pathogens present in the water system.

Rearing represents the greatest time and energy investment during the entire process of fish culture at an enhancement facility and as such, it is a period that requires care and attention to details that may seem relatively minor, but may well determine the overall health of the population.

4.12 Feed, Feed Storage, & Feeding Practices

Rationale: Proper storage and handling and distribution of fish feed is essential to maintain the nutritional value of the feed. Uneaten food generates an ammonia cloud as it breaks down and this can be detrimental to fish. Additionally, uneaten fish food generates suspended solids and these can lead to bacterial gill disease. If a maximum ration is fed to small fish, these constraints can be more restrictive than oxygen criteria. That is, more water is required to flush away particulates than is required to maintain dissolved oxygen levels. Low concentrations can be equally detrimental if sustained for long exposure periods. This SOP addresses section *2.1.2* of the General Principles of Fish Health Management. The goals of this SOP are to ensure that feed is stored in a manner that ensures its nutritional value is maintained and that feed is distributed appropriately to fish.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

4.12.1 ***Feed Storage***

Feed should be stored in secure buildings (i.e. freezer) such that wildlife is excluded and feed is kept protected from extremes of heat, light and humidity. Dry feeds should be stored at temperatures <20^{o}C and humidity <75%.

Windows and doors to feed storage buildings are to be kept closed to exclude pests.

Feed for immediate use and feed in feeders should be similarly protected in sealed top containers to protect it from humidity and light, and should be replaced frequently with feed from storage.

4.12.2 ***Feed***

Feed should be obtained from a feed mill that has been inspected by the CFIA. (EWOS and Skretting)

Feed bags should be labeled with the date of manufacture and guaranteed analysis information.

Feed should be rotated so that newer lots of feed on site are fed out last and any spilled feed is cleaned up immediately.

Feed buckets should be cleaned, disinfected and put away after use.

Medicated feed must to be clearly identified and used immediately. (See *Medicated Feed: Storage, Handling, and Feeding*)

4.12.3 ***Feeding Practices***

Fish should be fed at appropriate intervals with a nutritionally adequate feed. Feeding and feed size-sorting should be optimized to ensure all fish have the opportunity to feed.

Fish should be observed regularly during feeding to determine if they are responding as expected and if the volume of ration is sufficient or if overfeeding is occurring.

Overfeeding should be avoided due to its effects on water quality and the stimulation of potentially harmful bacterial and fungal growth.

When automated feeders are used, the equipment should be serviced regularly and the rate of intake of the fish checked frequently.

Failure to begin feeding or to acquire a sufficient amount of food is considered a major cause of death of larval fish. In the event of food refusal or failure to gain weight (as determined by routine bulk sampling of newly ponded alevins), Fish Health Management, Support Biologists (Brian Anderson is the lead on food related issues), the Veterinarian and the feed manufacturer should be informed.

Note: if a feed-related problem is suspected, a sub-sample of food from the lot in question should be bagged, labeled and frozen in case analysis is indicated.

Details of the Operating Procedure:

Feed is stored in the walk-in cooler. Feed is also stored in keeper building #3. Smaller allotments (approximately 15 bags) are kept in the feed sheds outside of these locations for daily feeding.

Feed buckets are cleaned and disinfected on a weekly basis, or more frequently if required.

The majority of feed at the main facility is delivered via the motorized Belrooke blower feeder.

Feed delivery at the lake site is predominantly by hand, but solar powered automatic feeders may be used on occasion.

Belt feeders are used in the circular containers and in the Capilano troughs. Some hand feeding is performed on these containers to ensure that fish are being observed on a regular basis.

Automatic Neilsen hopper feeders are used for delivering feed to ponds 9-12. Two feeders are installed on each pond when in use.

If feed is observed on the bottom of a rearing container, the number of feedings, or the duration of each feeding will be reduced.

First feeding:

Chum are fed as soon as they exit the keeper channels as the ponding process may be of up to 2-3 weeks in duration before the keeper is completely free of fry.

Individual groups of fry in Heath trays may be ponded over the course of one week before the entire group is transferred. Fry are not offered first feed until all the fry have been ponded. First feed will be offered 24 hours following the point when all of the fry have been transferred to the rearing container.

If fish appear to be refusing feed, the reasons will be investigated. The water source will be examined for changes in temperature, dissolved oxygen and flow rates. Mortality patterns will be examined for changes. If the problem does not appear to be environmental, other facilities will be contacted to determine if other facilities are experiencing similar difficulties.

4.12.4 *Set up and operation of feeders on ponds 9, 10, 11 & 12*

1. Install feeders in assigned locations
2. Go to control panel and perform the following steps : [panel located at the South end Bld. # 3]
    a. Turn on power to feeders to insure operational. [toggle switch on RH side of panel]
    b. Adjust daily clock for start, stop times over a 24 HR s. [clock located inside the panel]
    c. Set HR clock for the frequency of feeding in minutes [HR clock located on the RH face of panel]
    d. Set second clock for the length of time for each feeding. [Second clock located on the LH face of panel]
3. Use gauge to verify gates are set to proper opening. [gauge is a piece of ¼ * ½ flat bar 8" long]
4. Load feeders with required food
5. Turn on power
6. Check to verify feeders are working and the proper amount is being supplied to the fry.

Forms & Records:

Lake Feeding Sheet.xls

Stauffers Growth.XLS

Food Conversion

Food Order.xls

References:

Medicated Feed: Storage, Handling, and Feeding

EWOS Food Size Guidelines

4.13 Individual Length/Weight and Bulk Weight Sampling Protocols

Rationale:. Juvenile length-weight sampling is a random, unbiased method used to confirm and monitor fish development. Juvenile growth as well as environmental conditions will determine the ration and rate at which the juveniles will be fed during a rearing program.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Husbandry records should be reviewed to ensure no sign of disease within the population to be sampled. Time held off-feed should be confirmed. All equipment should be assembled and confirmed in good working order before starting.

Fish should be crowded toward the inflow end of the rearing container. Dipnet samples should be taken from various areas/depths of the crowded fish and placed into a large sample tote. This sampling protocol hopes to achieve a representative sample of the entire population.

During bulk sampling, a subsample of fish, totalling ~100 fish should be taken from this group and transferred to a smaller sample bucket (~5 L), and transported to the weigh station with minimum disturbance. Fish should be netted out and drained of excess water before being placed into a tared container of water. The weight should be recorded and fish should be netted out and counted back into a recovery container or back into their enclosure. Repeat samples may be done on each rearing unit.

During length weight sampling, a sub-sample, totalling ~ 50 fish should be taken from this group and transferred to a smaller sample bucket (~5 L), and transported to the weigh station with minimum disturbance. Anaesthetized fish should be gently placed on the measuring board; length (standard, fork or total as directed) should be recorded. The fish are then transferred to the balance for weight measurements and/or placed into a recovery bath. Repeat samples, to a total of 3 per rearing unit, may be done.

Anaesthesia and *Fish Handling Procedures* SOP guidelines should be followed. Anaesthetic baths should be changed between rearing containers, or if time till anaesthesia lengthens or if the bath temperatures differ >2^{o}C from that of the rearing container. Anaesthetic baths must be disposed of in accordance with local waste management regulations.

Details of the Operating Procedure:

4.13.1 ***Bulk Sampling:***

Bulk sampling is used to estimate the weight of the entire population of a rearing container during juvenile development. Juvenile growth as well as environmental conditions will determine the ration and rate at which the juveniles will be fed. Lack of expected gain can be the first indicator of a feed

quality, disease or water quality issue. Additionally, sampling gives the opportunity to visually inspect juvenile fish for clinical signs of disease.

Bulk sampling is performed approximately once a week for determination of appropriate feeding rates and feed sizes.

Equipment list:

Person/equipment to crowd fish
Person to sample fish
Appropriate sized sample dipnet
Smaller sub-sample net
Balance
One appropriate sized tote
Sample pails (~5L)
Notepad with waterproof paper and pencil

Note: Fish should not have been fed within the 18-24 hours prior to sampling

1. Assemble all equipment
2. Locate an area of the pond where there is a large concentration of fish
3. Plunge a dipnet into the centre of the populated area and collect a random sample from the group
4. Using a smaller net, dip out a random sample of the fish and place them into a 20L pail containing water
5. Place a small (1-3 L) container containing water on the scale
6. Set another pail containing water at the outflow of the flume
7. Tare the scale to zero it
8. Dip out a subsample of the fish, dewater them for a moment and then pour them into the tared container
9. Record the weight
10. Pour the fish slowly into the flume and, using a tally counter, count them as they pass through it into the second container
11. Place the fish back into the pond from which they were collected
12. Approximately 300-400 fish should be counted for each pond bulk sampling
13. Clean and put away all equipment after the procedure

$$\textbf{Mean Weight} = \frac{\textbf{Total Weight of Sample}}{\textbf{Number of Juveniles}}$$

4.13.2 ***Length/Weight sampling:***

Juvenile length-weight sampling is a random unbiased method used to confirm and monitor fish development. Accurate size information is a valuable tool to help a manager coordinate release date and size targets in an attempt to mimic the natural life stages of wild juvenile fish. Juvenile growth, as well as environmental conditions, will determine the ration and rate at which the juveniles will be fed during a rearing program.

Length-weight sampling is performed at ponding, at midway through rearing and a few days prior to release

Equipment list:

Person/equipment to crowd fish
Person to sample fish
Large sample dip-net and tote
Smaller sub-sample net and labeled sample pails (5L)
Anaesthetic equipment (drug, buffer, airstones, dedicated basin, thermometer, etc.)
Balance
Measuring board or ruler
Gloves
Notepad with waterproof paper and pencil or computer

Notes:

- Fish should not have been fed in the last 18 - 24 hours
- Fish may or may not be crowded for sampling
- Be gentle with the fish and handle as little as possible.
- Monitor closely for signs of stress.
- Work as quickly and as efficiently as possible.
- Record all data and report any unusual behaviour or physical characteristics.
- **Disinfect all sampling apparatus**

1. Assemble all equipment
2. Prepare the anaesthetic bath ensuring oxygen is being delivered to it
    - Anaesthetic = 0.25 g MS-222 + 5 L water + 1 Tablespoon Buffer (Baking soda)
3. If not crowding fish for sampling
    - Locate an area of the pond where there is a large concentration of fish. Feed may be applied to generate a “feeding boil” from which fish may be collected
    - Plunge a dipnet into the centre of the populated area and collect a random sample from the group
4. If crowding fish for collection
    - Insert a screen downstream of the inflow to reduce injury to the fish from the turbulence
    - crowd fish towards the pond/container inflow

- if the water is clear, crowd fish to the point where the bottom of the screen is not visible
- if the water is cloudy, crowd fish to the point where they seem "unhappy" about the situation.
- plunge the dip net into the concentration of fish

5. Remove 3 to 5 nets of fish from the population at different points in the pond and place in a large bucket containing water
6. Move the container of fish into the lab where sampling will take place
7. Set the bucket of fish in the sink
8. Place a container with water ready on a scale (remember to tare scale to 0.00)
9. Place a recovery bucket nearby and ensure that oxygen is being delivered to the container
10. Transfer a small number of fish (10-30) into the anaesthetic bath
11. Wait until the majority of the fish show signs of inactivity before sampling
12. Using a small dipnet, remove fish individually and place on the measuring board
13. Measure length of fish in mm (nose to fork in tail)
14. Record the measurement in the data sheet
15. Use a sponge to remove the excess water from the fish
16. put the fish into the container, record weight in grams
17. Repeat this process until a minimum of 150 fish have been sampled from the pond
18. Place the fish into the tared container
19. Record the weight
20. Remove the fish and place it in the recovery bucket
21. Tare the scale before placing the next fish in the container
22. When each sub-sample of fish have recovered, return them to the pond from which they were collected
23. To determine the Condition Coefficient (KC) :

    ((Weight x 1,000) divided by (Length x Length x Length)) * 1,000
24. Clean and put away all equipment after the procedure
25. Enter data into EnPro

Forms & Records:


Juvenile Sampling.xls


Loading rates

EnPro

References:

Fish Handling Procedures
Anaesthesia
Equipment disinfection
Growth prediction tables should be available for comparison

4.14 Fish Handling Procedures

Rationale: Handling of fish must be done in a manner that minimizes stress and injury and minimizes the risk of escape. This SOP addresses section *2.1.3* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that fish are handled in as stress free a manner as possible and to ensure that the risk of fish injury or escape is minimized.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Feed should be withheld from fish for a minimum of 12 hrs and a maximum of 72 hrs prior to handling. Determination of the time off food includes consideration of fish size, diet, water temperature and existing knowledge about gut emptying time (gut emptying times are longer for larger fish and colder temperatures).

The crowding of fish should occur for the least amount of time possible to reduce injury and minimize stress.

Materials used in handling fish should have smooth surfaces and be designed to minimize injury to the fish.

Fish must be adequately supported when out of water and must not be handled solely by the tail (if expected to survive the procedure) which can damage the vertebrae.

Time out of water should be minimized.

Prolonged physical restraint of un-sedated fish should be avoided due to possible damage to the skin and mucus coat. These barriers are critical to protect fish from osmotic stress and infectious agents. Anaesthetics or sedative agents can be used to minimize the stress and injury risk associated with handling procedures like vaccinating, marking, grading and sampling (see *Anaesthesia*).

When fish are handled out of water, anything they contact should be kept wet to minimize abrasions and loss of mucus. Mucus replacements/protectants (i.e. Vidalife™) can be used on handling equipment, within anaesthetic baths or transport water to protect the mucus coat of the fish.

Water quality (particularly oxygen and temperature) should be monitored before handling fish. Depending on the procedure, water quality may be measured throughout the handling procedure.

Dip net loads should not contain excessive numbers of fish. Pipes used to move fish should be smooth inside with no sharp bends, or excessive or inadequate water flow.

After handling, fish should be examined for signs of injury or scale loss. Fish should be monitored closely for several weeks following the handling episode to allow rapid detection of signs of injury or disease.

Details of the Operating Procedure:

Holding units are made of materials that limit injuries to fish. During design and fabrication of fish holding units the materials chosen are properly reviewed and screened before being purchased. Incubation holding areas are made of plastic or aluminum. Small circular tanks are made of plastic. Raceways are made of concrete. Coho channels are made of asphalt and lined with plastic.
If greater than normal mortalities have been observed, handling procedures will be cancelled and the issue will be investigated.

Fish are starved prior to handling procedures a minimum of 24 hours.

When fish are taken out of the water for any length of time they need to be properly supported; they must not be subjected to crushing due to the weight of fish above them. When fish are removed from rearing containers it is either by dip net or fish pump. Nets are made of soft mesh to help prevent scale loss or other physical trauma. When fish are pumped, they are out of the water for a very brief period of time. They are transported into the machine in water and on the metal surface for de-watering for < 1 minute before being put into a holding tank with water. There is generally a lower possibility of being crushed in the fish pump than in the net as the depth of fish on the metal surface is generally <2.5cm of fish.

4.14.1 ***Seining procedures***

See *Broodstock Handling* methods

4.14.2 ***Handling procedures during marking***

Coho and steelhead are marked with an adipose clip; Chinook are marked with an adipose clip and coded wire tag. Chum and Chinook otoliths are marked by temperature fluctuations during incubation. Fish are closely monitored during and after handling for signs of scale loss and skin abrasions. When fish are marked they are visually assessed after the marking process and assessed daily, once they have been returned to their rearing containers. Mortality rates are monitored more closely after fish have been handled for any reason.

Generally, other than sampling and marking of juveniles, when fish are moved it is to be released thus a post release assessment is not feasible.

See *Marking Fish*

4.14.3 ***Collection for Sampling***

1. If not crowding fish for sampling
    - Locate an area of the pond where there is a large concentration of fish. Feed may be applied to generate a “feeding boil” from which fish may be collected
    - Plunge a dipnet into the centre of the populated area and collect a random sample from the group

2. If crowding fish for collection
    - Insert a screen downstream of the inflow to reduce injury to the fish from the turbulence
    - crowd fish towards the pond/container inflow
    - If the water is clear, crowd fish to the point where the bottom of the screen is not visible
    - If the water is cloudy, crowd fish to the point where they seem "unhappy" about the situation.
    - Plunge the dip net into the concentration of fish and collect a random sample from the concentrated fish

Forms & Records:

References:

http://www.syndel.com/handling/vidalife_info_sheet.htm

Anaesthesia

RiverSeining STEPS

4.15 Marking Fish

Rationale: Fish are marked for identification purposes. The procedure should be done in a manner that causes minimal injury and stress to the fish. This SOP addresses section *2.1.3.2* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that fish are marked properly with minimal stress or injury. Methods of marking should not negatively affect productivity or survival unless they are part of an institutional approved research protocol.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Crowding time should be minimized and equipment used to handle fish should have smooth surfaces and be designed to minimize injury to the fish (see *Fish Handling Procedures*).

Fish may be collected in dip nets or pumped to the marking location. When dip nets are used, the mesh should be soft and knotless. Net loads of fish should not contain excessive numbers of fish. No more than 1/3 of the net volume should contain fish to avoid crushing fish at the bottom of the load.

Time out of water must be minimized and if the transport to the marking station exceeds more than a few seconds the fish should be transported in a bucket or tote containing water.

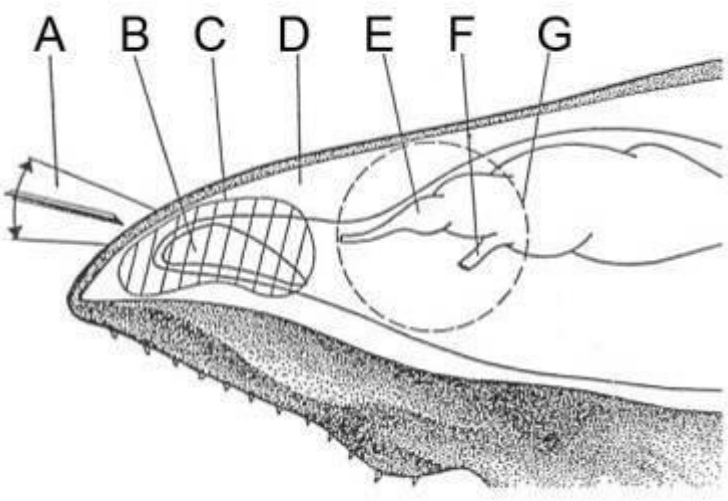
Fish should be anaesthetized for marking procedures (see *Anaesthesia*).

All equipment used for marking procedures should be cleaned and disinfected prior to use and between groups of fish (defined units). This will decrease the risk of pathogen spread between fish (see *Equipment disinfection*).

For ***adipose fin clipping***, the adipose fin is removed to serve as a quick visual identifier of hatchery origin fish. Proper removal of the adipose fin provides a life-long mark which is not considered to adversely affect the health, behaviour or social interactions of the fish.

Scissors are used to remove the fin and fish are placed in anaesthetic-free water for recovery. Scissors should be disinfected periodically and replaced as necessary to ensure sharp cutting edges are used.

For ***coded wire tags***, the tag is implanted in the rostral cartilage and the fish should be immediately placed into anaesthetic–free water for recovery. Periodically, fish should be humanely sacrificed to confirm proper placement of the tag.



Typical Coded Wire Tag Placement

A. Usual range of tagging needle angles
B. Muscle, adipose and fibrous tissue
C. Tag target area
D. Cartilage
E. Olfactory lobe and nerve
F. Optic nerve
G. Position of eye

Otolith marking is performed during incubation by varying the temperatures that the fertilized eggs or alevins are exposed to. A specified temperature change over a predetermined amount of time creates a life-long mark with no known adverse effects on hatching success or subsequent fish survival, health, behaviour or social interactions. However, this mark can only be judged through lethal means and is therefore more of a tool for stock assessment purposes. As the samples must be sent out for analysis, and this analysis can take a considerable amount of time, it should not be considered to be a visual tool for hatchery staff.

Details of the Operating Procedure:

4.15.1 ***Fin clipping***

A proportion (determined by the sampling plan provided by Headquarters) of coho are adipose clipped each year

Fry are marked at approximately 1.5 g. Outplants are marked approximately 2-4 weeks prior to release. Yearlings are marked year prior to release.

The fish are starved for 24 – 48 hrs prior to marking

1. Assemble all required equipment and set up the marking and tagging stations
2. Set up holding containers and ensure the water contained within is cool and well oxygenated
3. Set up the TMS anaesthesia bath. Ensure that the system is oxygenated.
4. Fill a Rubbermaid pail with water and place it on the 6 wheeled cart
5. Net fish out of their rearing containers into a large Rubbermaid pail with volume lines. Volume lines are in 2kg displacement increments. Collect the required number of fish based on weight/displacement
6. Transport the container to the tagging room
7. Move the fish into holding containers using a dip net
8. Ensure that densities in the holding containers is at densities similar to or less than those found in the rearing containers
9. From the holding containers small numbers of fish are dip netted into 3 litre anaesthetic baths to which a predetermined amount of buffered TMS stock solution has been added
10. The fish are transferred to a 3 litre freshwater basin for marking. Fish marking requires excellent dexterity and as such the markers do not wear gloves. Therefore the transfer of the fish from anaesthetic to freshwater prevents exposure of handlers to the drug.
11. Keep total anaesthesia exposure time to < 2 minutes; staff levels must be maintained at a sufficient number to minimize anaesthetic time
12. Using a pair of micro surgical iris scissors remove the adipose fin completely
13. The coded wire tagging protocol from the "Juvenile Marking Manual" (1990) by T.L. Nichols and J.E. Hillaby is used
14. Count each fish on the tally counter during the marking process
15. *Place the clipped fish into the flume which carries it to the recovery trough. The fish are returned to their container via hose*
16. *Clean and disinfect all equipment using Hyperox following marking.*

4.15.2 ***Otolith marking***

Mass marking of hatchery stocks is highly desirable for a variety of reasons and can be easily accomplished by thermal otolith marking. This can be done with fish at almost any time during early development and is primarily applied at the post eyed to pre ponding period, excluding hatch. Sudden changes from ambient water source to cold supply for 24 hours and then switching back. A minimum drop of 1.5^{o}C will create optically dense (dark rings) on the otolith. Discreet thermal marks for different stocks can be applied in a well-designed simple marking schedule during incubation.

Otolith marking is performed during post-hatch incubation over an approximately 15 day period.

Sarita Chinook receives a 3:2:3 mark
Nitinat Chinook receives a 2:3:2 mark

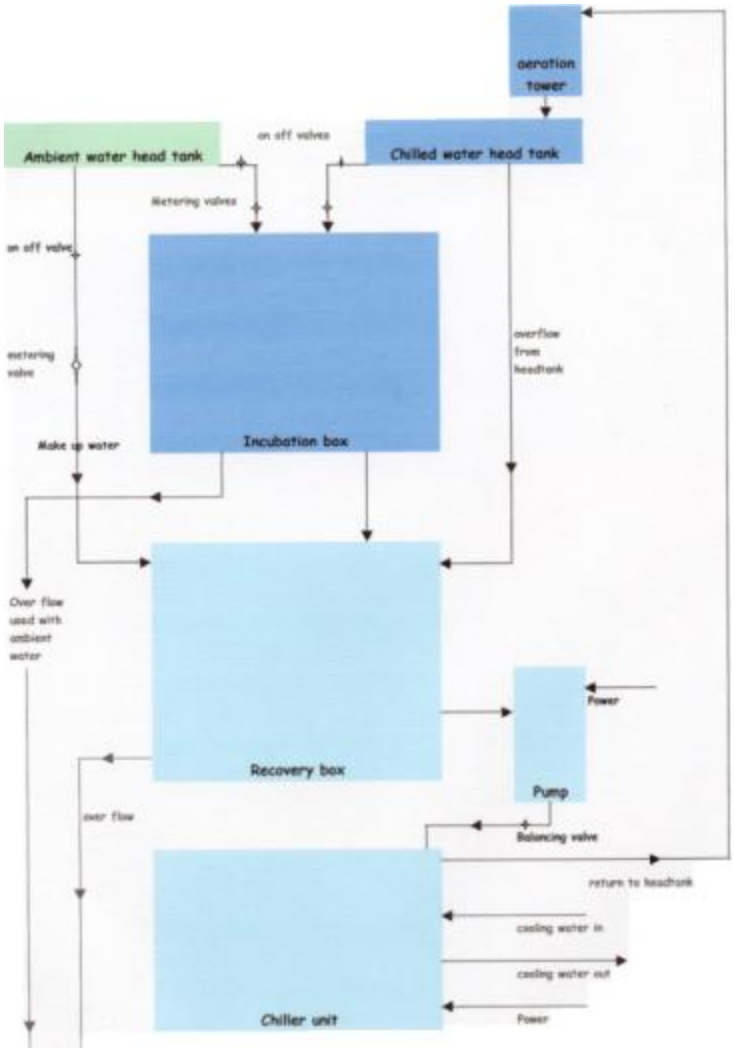
4.15.2.1 Chilled Cycle – Thermal marking– "Dark Rings"

1. *Turn off ambient water in head tank 3 by "swinging" (A2) supply to upright position*
2. *Turn on Chilled water at head tank 4, at valve (C2)*
3. *Leave catch trough under outflow of the incubator box until water level is 5-10 cm above screen diffuser of recovery box*
4. *Remove catch trough and install spill plate*
5. *Recovery box will fill with makeup water supply @ 10 L/min,*
6. *Monitor to confirm recovery box is filling*
7. *Record all actions on 2nd incubation sheet & otolith sheets.*

4.15.2.2 Ambient Cycle – Warm cycle - "Light Rings"

1. *Turn off chilled water at Head tank 4, by closing valve (C2)*
2. *Remove spill plate and install catch trough under "spill" at outlet of incubator box*
3. *Turn on ambient water (from Head tank 3) by swinging ambient supply to down position (A2)*
4. ***Do Not Turn Off Chiller or Pump**– this will ensure system is kept cool for next chilled cycle*
5. *Record all actions on 2nd incubation sheet.*

Chiller schematic



Forms & Records:

FINCODES.DOC Clipping.xls

References:

Equipment disinfection
Anaesthesia
Fish Handling Procedures

4.16 Juveniles-Health Observations

Rationale: Changes in physical condition and behaviour are good indicators of poor health and/or disease. Early detection is key to good disease management. This SOP addresses section *2.3.3* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that changes in physical condition and behaviour that may indicate poor fish health and/or disease are identified.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:
Fish are observed daily for signs of health, injury and disease.

Changes in behaviour (decreased feed response, decreased startle response, failure to evade capture, etc.) and physical condition (darkening in colour, failure to gain, external lesions, etc) are reported and recorded. Any change should be investigated and the causes identified and corrected.

Groups of fish are tracked from their incubation containers to their rearing containers using the ENPRO Juvenile Manual.

Biological records include: species, stock, length, weight, condition, and comments on appearance.

Groups of fish suspected of having a disease are sampled according to the Veterinarian's instructions for lab analysis.

Details of the Operating Procedure:
Fish are observed on a daily basis, during feeding, for signs of health, injury and disease. Observations of feeding behaviour, water quality etc, are noted. Feeding may be adjusted or withheld based on these observations.

Changes in behaviour (decreased feed response, decreased startle response, failure to evade capture, flashing, crowding to an area of the pond, position in the pond, etc.) and physical condition (darkening in colour, failure to gain, external lesions, etc) are reported to the Supervisor and recorded in the broodstock forms and the juvenile rearing records. Any change should be investigated and the causes identified and corrected.

Observations are recorded on the weekly container rearing/record sheets

The supervisor is notified of any suspected problems. Observations are discussed each week at the staff meetings, but unusual problems will be communicated immediately, Veterinarian will be contacted and fish may be sent to the pathology laboratory for diagnostics.

Forms & Records:
References*:*
Diagnostic Sampling protocols

Release

4.17 Pre-Release or Transfer Disease Risk Assessment

Rationale: Transferring fish between facilities and/or watershed represents a potentially serious breach in biosecurity. The risks are deemed acceptable in situations where the conservation concerns and or marginal water availability or quality make enhancing stocks at a single site impossible. The pre-transfer disease risk assessment helps to inform the Fish Health Management Team of the relative risks of moving fish between sites and to identify mitigating factors to lower the risks associated with animal transfers. The goal of this SOP is to ensure that the decision making process in the pre-release or pre-transfer disease risk assessment process has been appropriately detailed.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

With a long-standing established program involving annual fish transfers between two sites, with appropriate surveillance data collected and with historical knowledge in endemic disease issues in the two populations, the disease risk assessment may be relatively informal. Any such transfer program should be reviewed during the facility annual production planning process.

No sick fish should be transferred between sites or knowingly be released without a disease evaluation. Depopulation, treatment and release options should be reviewed on a case by case basis.

Where new programs are developed or in instances where the rearing population has suffered disease losses and treatment, the Veterinarian may request a sample of either healthy or moribund fish for disease prevalence estimation at least 2 weeks prior to transfer/release.

Assessment:

For each proposed transfer of fish the Fish Health Management Team should consider the following information:

- species, life stage, disease and treatment records
- location of receiving facility or watershed
- disease history of the current rearing facility
- history of pathogen surveillance within the population being moved
- history of pathogen surveillance and prevalence in the feral populations within in the receiving waters
- availability of post-release isolation or disease sampling and diagnostics

References:
Juvenile Release
Juveniles-Health Observations
Mortality Classification

4.18 Transporting Fish

Rationale: Transportation of fish is a complex and stressful event. Fish must be handled in a manner that protects their health, minimizes the length of the stressful event and mitigates risks to any fish at the receiving site. This SOP addresses section 2.1.3.3 of the General Principles of Fish Health Management. The goal of this SOP is to ensure that fish are transported in a manner that protects their health, the health of the fish at the receiving site and is done in accordance with all regulations.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

4.18.1 ***Prior to transport***

In consultation with a qualified fish health professional, the diagnostic and treatment history of any fish being moved should be reviewed prior to transport (see *Pre-Release or Transfer Disease Risk Assessment*). This includes mortality, diagnostic and treatment records and examination of a representative sample of dead fish and moribund fish within 10 days of transportation. Consideration should be given to any differences in the pattern of disease recipient area and the location which fish are being transferred.

Fish showing signs of illness or fish held under quarantine for any reason should not be moved.

All necessary permits/licenses should be in place

If fish are to be moved from fresh to salt water then the degree of smoltification may be assessed to ensure that fish are ready to be moved.

Fish should be taken off feed for at least 24 hrs prior to transportation.

All required equipment should be available and functioning.

Water used for transportation should be whichever available source poses the lowest risk for pathogen transfer. In freshwater sites, well water is preferable.

Fish should be crowded for the minimum time possible to allow collection into transport vessels via pumps or nets. Dip nets should not contain too many fish and pipes used for pumping fish should have smooth surfaces and minimal bends. Water flow during fish pumping must be neither excessive nor inadequate.

Fish transfer into the vehicles/vessels must be conducted in the least stressful manner possible. Fish should be handled in a manner that minimizes skin damage or other trauma and leaves fish out of the water for as little time as possible. When fish are handled out of water, the equipment used to handle them should be kept wet to minimize trauma.

Options to help lower the stress of transport and reduce the risk of injury to the fish include the use of mucus protectants (e.g. Vidalife™), the addition of 3-5 ppt of salt to the transport freshwater or the addition of a sedative (e.g. Aquacalm™ - not to be used when fish are to be released due to mandatory withdrawal period).

4.18.2 ***During Transport***

Water quality must be maintained at all times during transportation. Transport vessels should be equipped with supplemental oxygen tanks and air stones. Oxygen levels should be checked and maintained throughout the transport procedures; frequency of monitoring will vary with the specifics of each transport, as a generality, oxygen should be measured every 30 minutes. Fish should be visually monitored regularly (every 30 minutes) during transport to ensure they are behaving in an expected manner.

Other water quality parameters which may be measured include temperature, pH and ammonia. The parameter measured and the frequency should be determined by the duration of the transport.

A contingency plan needs to be in place to ensure that if the transportation is delayed water quality can be maintained. Ice packs or ice made with confirmed non-chlorinated water may be used to keep transport water temperatures down. Ice slush is safer for the fish than chunks of ice.

Water quality in the transport vessel should be matched to that in the receiving water wherever possible. A slow acclimation to new water quality is preferable to dramatic changes. Water temperatures should ideally not differ by more than $2^{o}C$ and dissolved oxygen levels should be equivalent so that gill tissue is not damaged.

Fish should be released into the receiving waters in a careful manner. Locations of all groups should be noted in the records.

4.18.3 ***After transport***

Vehicles and vessels and equipment used in transport should be cleaned and disinfected after use.

New arrivals to a site should be isolated from fish already on site. Fish should be monitored closely after transportation for signs of illness or trauma. Mortality rates should be calculated at least twice weekly after transport and all fresh mortalities should be examined for the next two weeks to ensure that a pathogen has not been brought unintentionally onto the site. .

<u>Details of the Operating Procedure</u>:

4.18.4 *Fish transport check list*

- □ Notify maintenance of impending transport at least 48 hours in advance when possible
- □ Check dumpsite before transport.
- □ Ensure that the transport hose is long enough
- □ Determine tank load(s), (kg amount of fry to be loaded).
- □ Have an area map of transport route

- □ Collect the clipboard with necessary transport data sheets
- □ Add 1 kg of salt to each tank to be used
- □ Fill the tank(s) with water to the site gauge mark required
- □ Ensure seine and dip nets are ready at the fish loading site
- □ Calibrate the DO meters and ensure they are set to read saturation percentage
- □ Install DO meters in tanks
- □ Charge the oxygen levels in the tanks to 19.0 ppm, and turn the oxygen flow off
- □ When using the 5 ton with 3 tanks, number the tanks #1, #2 and #3, from the front to the rear of the truck. Make this the standard for all fry transports

4.18.4.1 Prior to movement of fish:

The day prior to moving fish, all associated equipment, including trucks, tanks, pond nets, fish pump, oxygen regulator, flow meters, airstones O_2 meters should be checked, disinfected and calibrated.

On short 'hauls' (<1 hour; i.e. from hatchery to lake site net pens), chum and Chinook are starved for at least 24 hours prior to transport. Most of our juvenile movements and outplants to the lake are multi-day events, so total time "off food" can be 48 hours to 72 hours . On longer hauls (>1 hour), fish are routinely starved a minimum of 48 hours.

4.18.4.2 Movement of Fish:

4.18.4.2.1 Groups transported to and from Nitinat

Juveniles:- March^{t} – June 15th

- Hatchery site to Nitinat lake net pens. chum [0.6gm] and Chinook [min 2.5gm] < 1hr/trip;)
- Hatchery site to upper watershed (>1hr/trip coho [3gm], steelhead [3gm])
- Hatchery site to Sarita river (Chinook [min 4.5gm] >1 hr./trip)
- Hatchery site to Sooke Basin (Chinook [min 4.5gm.] >1 hr/trip)

Broodstock:- September to November

- Nitinat river to hatchery (Chinook and coho adults <1 hr/trip
- Nitinat lake to hatchery (chum adults) <1 hr/trip

Transport and receiving water are normally very close in temperatures. If receiving water temperatures are greater than 5°C different from transport water, the fish will be acclimated prior to transfer. Transport during the spring program occurs from March 1 – May 31st; fry transport trips rarely exceeds more than a 3°C rise in the tank over a 3 hour period.

At Nitinat, fish from juvenile to adult are transported in tanks filled with 7^{o}C - 8^{o}C well water and charged to 20-25 ppm oxygen. On trips >1h salt is generally added at 5ppt - 10ppt prior to filling to ensure it is dissolved. Once required dissolved oxygen levels are reached, the flow meters are 'throttled' back. The truck is then moved into position and the tank water levels adjusted to pre set fry density levels of 0.1 - 0.125kg/L. A slime replacing agent 'PolyAqua' is generally added to transports of > 1h in duration. All juvenile salmon transports are based on volume displacements - One kilogram of fish will displace ~1 L of water.

Fish are gently crowded into a pond net liner and toward the pump intake. While this is going on, the pump is started and priming begins. The flow meters are set at 4 L/min for < 3g fish and at 2 L/min for >3g fish. Once priming is complete and fish movement begins, pump speed and fish crowding is adjusted to allow for a "comfortable" movement of fish from pond to truck. Intake hose from pond to pump hopper is transparent and fish movement through the system into the tank is observed closely. This entire process works well with a minimum of 4 people. Tanks are generally loaded in 5-8 minutes. This is dependant on fish size. Chum (at 0.6g) take more time to load than Chinook (at 6 g).

At this point, final visual checks of the fish and equipment are made.

On short haul trips, (i.e. to Nitinat lake net pen site), only the driver is present in the truck. The driver makes radio contact with the lake crew (2 people) once the truck leaves the hatchery site. Radio contact is made again 5 minutes prior to arrival at the lake. Once contact is made, one person at the lake arrives at the discharge site, starts the "flushing pump" and waits for the truck. This person acts as a "swamper", and ensures that all the 'small details' are looked after, before during and after the fry are relocated to the net pens some 200-ft. away. The driver manages the truck by ensuring it is in the correct position, opening all tank lids, visually inspecting and assessing the fish in all tanks and monitoring D.O.s, before giving the OK to release. Discharge DO levels should be at 10ppm – 15ppm. This helps reduce some of the "skittering" seen upon the surface of the net pens when oxygen (gas pressures) are too high.

Once all the fish have been released, the flushing water supply is connected to the discharge hose and run for 15-20 minutes to clear any fish that remain in the line.

On longer hauls all procedures are similar to the short trips, with the exception that there are a minimum of 2 people in the transport truck and regular inspection stops are done every hour. The Sooke basin trip takes up to 3 hours and can use up to 400' of transport hose.

4.18.4.3 Broodstock Capture and Transport

During the fall program up to 80% of ***Nitinat Chinook*** broodstock are annually transported from "Red Rock" pool 15 minute away on the Nitinat River. Beach seine sets of up to 2000 – 3000 adults have been made and require 10-15 people, handling up to 1000 adults at an average of 25 fish per transport trip over a 6-7 hour period. Once the set is made all surplus (excess to the days needs) fish are released. There is a constant flow of truck and fish loading activity, where the emphasis is on safety, and minimisation of stress to fish production efficiency.

At the hatchery, the driver quickly discharges the adults via a slide into the deep ($225m^3$) ponds. Three trucks with single 1400 litre tanks can make 2 trips/hour.

Water quality parameters are monitored as described for juvenile fish (above).

Transport of ***Sarita Chinook*** requires substantially more time and "delicate" handling. They are caught in a small beach seine in smaller numbers and in harder to access areas on this river. Chinook caught in the beach seine are counted and released back to the river or are held in adult (max. 2 per bag) handling bags. Maximum tank capacity for these trips is 15 adults. Site specific conditions for the Sarita River requires different "transport rules". September air and water temperatures are usually $15+^{o}C$. Keeping transport water cool with ice, addition of a slime rebuilder (PolyAqua at 100ppm) to

reduce infection and fungus and lowering of tank densities have helped minimise transport loss and pre-spawn mortalities at the hatchery. Gamete quality has also markedly improved when the adults are cared for in this way.

Transportation of ***chum broodstock*** from our lake operations is an "opportunistic" process. Where Nitinat Chinook loads can be 2 –2.5 kg/L, chum transport can be nearly double at 4- 5 kg per litre or (40x –50x our fry densities. Broodstock caught in a commercial seine are held for 1 week in net liners until numbers and a "window of opportunity" presents itself to allow for the eventual transport of up 10,000 - 15,000 adults or ~120 chum/1400 L tank) to be transported over a 3 week period.

4.18.4.4 After Transport

Transportation equipment is cleaned after use. Where organic matter has built up it will be flushed out with clean water. If possible the transport tanks are allowed to dry between uses. Transport tanks will be disinfected if large numbers of fish died during transport (>1% mortality)

4.18.5 *Pumping fry from all shallow ponds from a one time set:*

1. Set up pump below pond #8 in the release-way
2. Place stop logs at the East end of the release way to desired depth.
3. Place a screen in the release way at the East end of pond 8
4. Place stop logs at the West end of the pond being pumped [This will divert the water from remaining ponds to the Little Nitinat release site]
5. Install newly designed and manufactured basket net [as used in the concrete ponds , it may prove out that this net is not needed]
6. Pull the V notch and stop logs from the pond being pumped
7. Crowd (if necessary) the fry from the pond to the release way
8. Pump the fry from the release way to the transport tanks
9. Adjust the height of the water in the pumping section of the release-way as required by removing stop-logs at the East end of the release-way
10. When down to the last remaining fry use the basket net if required
11. When one pond is finished readjust stop logs for the next pond to be pumped

Forms & Records

Transport Info Form.dot

TRANSPORT.XLS

References:

Pre-Release or Transfer Disease Risk Assessment

4.19 Juvenile Release

Rationale: Fish are to be released in good health to minimize the transfer of pathogens to wild fish. The timing of release is also important to reduce stress and maximize survival of released fish. This SOP addresses section *2.5* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that the impact of hatchery fish on wild fish is minimized, to reduce stress and to maximize survival of fish being released.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

Details of the Operating Procedure:

Fish should be returned to their natal streams. Release sites and rivers should be checked for adequate flow, level and temperature, prior to release. Fish should not be released if there is a potential for extreme conditions in the river.

Other watershed users should be made aware of the time and date of the fish release if appropriate

Fish may be given a salt-water challenge test before release to determine smolt readiness.

Fish designated for the watershed should be checked for health condition prior to release.

Fish may not be fed on the release day, and may be sampled the morning of the release day. Fish that have not left within 2 weeks of screen removal may be forced to leave.

All equipment (tanks, pumps, hoses, aerator, air stones, etc) should be pre-checked to ensure good working order.

Details of the Operating Procedure:

Review the **Fish transport check list** *prior to moving fish*

Determination of time, weight and size of fish at release is based on targets set out at the beginning of the production year.

4.19.1 ***Coho Yearlings***

Coho are released directly from the facility to the river. Remove the gate at the outflow and allow the juveniles to leave volitionally. Remove the stop logs incrementally over 3-4 days. Generally approximately 90% of the fish will leave during this period. The remaining 10% remaining in the channel after 3-4 days will be forced out by lowering the water level and reducing the water flow.

Always leave one or two stop logs in place to protect the yearlings from predation

Any fish remaining when the ponds are to be emptied are swept out with a broom.

4.19.2 ***River release***

Check water conditions prior to planning a juvenile release offsite. When the target date is nearing, check the rivers to determine if flows are too great for release. If the levels are up and the water is turbid, it is a good time for release and fish will be better protected against predation.

4.19.3 ***Truck Releases***

If fish are to be transported long distances, add VidaLife to the transport water to reduce stress on fish

1. *On arrival at a release site, measure and record water temperature of both the receiving water and the transport water*
2. *Check the oxygen water of both the receiving and the transport waters*
3. *If the oxygen level in the transport water is higher than that of the receiving waters, reduce the oxygen flow and allow the fish to deplete the transport water of oxygen until the waters are relatively similar.*
4. *Open the transport tank lid and turn the oxygen delivery system off*
5. *Remove the airstone from the transport tank so that it does not become stuck in the outflow or result in damage to fish while releasing*
6. *Hook up the release hose*
7. *Fill several buckets with water for flushing out any fish remaining*
8. *Run the release hose out to an appropriate release location away from any large rocks or other potentially damaging objects (or into the net pen)*
9. *Ensure one person has a secure grip on the release hose and directs it in a slightly upward direction across the top of the receiving water*
10. *Open the release valve and release the fish*
11. *When the water and fish have been released, flush any remaining fish out of the tank/hose by pouring several buckets of water into the transport tank*
12. *Walk hose out to ensure all fish are safely out of the hose*
13. *Roll up the release hose and replace all equipment into transport truck*
14. *Return to the hatchery and disinfect any equipment, if required*
15. *Return all equipment to its proper storage location*
16. *Remove transport tank from truck if no further transports are planned*

4.19.4 ***Net Pen Releases***

Contact marine site users and notify them that the pens are to be installed (e.g. Poet's Nook). Ensure volunteer groups are aware of transfers. Contact forestry companies to ensure necessary gates are open.

Two weeks prior to transfer, set up the net pen frames at the holding site.

The day of transfer, install the nets onto the pen frames. This reduces fouling buildup.

900 kgs of fish are stocked into the 40' x 40' net pens. Fry will be kept in the pens until the biomass increases to 1800 kg. This will be approximately three weeks of holding but this will vary depending on conditions, feed availability etc.

Perform daily water quality checks in the net pens to ensure the water conditions are not deteriorating.

Fish are sampled one week prior to release.

When the fish are ready for release, unhook three sides of the net and pull the liner up carefully, allowing the fish to swim free.

Fish are released into the pen as outlined above in Truck Releases.

Forms & Records:

References*:*
Pre-Release or Transfer Disease Risk Assessment
Transporting Fish

Mortalities and Responses

4.20 Mortality Collection and Disposal

Rationale: The presence of mortalities in the rearing area can contribute to horizontal transmission of disease, attraction of predators and have negative effects on water quality and hygiene in the environment. This SOP addresses section *2.3.3.2* of the General Principles of Fish Health Management.

The goal of this SOP is to ensure timely removal of dead fish from the rearing environment; to ensure that dead fish are collected, stored and then disposed of in a manner that decreases predator attraction and pathogen spread and ensure hygiene and water quality protection.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Staff should take hygienic precautions to protect their own health; this includes wearing gloves while handling dead fish and washing hands after mort picking.

Mortalities should be picked from the youngest and healthiest groups of fish first. The same sequence for moving between tanks should be followed for routine sample collections.

Each holding unit should have its own equipment (nets and buckets) for mortality removal. Equipment used to remove mortalities should be cleaned, disinfected and dried between uses.

All dead and moribund fish should be removed from holding units on a daily basis. Moribund fish should be humanely euthanized prior to disposal (see *Euthanasia*).

Where feasible, separation of staff occurs so that staff picking morts are not feeding fish or cleaning holding units on the same day. Footbaths and hand wash stations should be used prior to returning to fish holding areas.

Mortalities should be counted and classified as they are collected. (*Mortality Classification*)

In the event of unexpectedly high morbidity or mortality rates, the frequency of mort collection may be increased. If daily mortalities exceed 0.5%, fish health management should be notified and the veterinarian consulted. (see *Outbreak Response* and *Outbreak – Disinfection Protocols* SOPs)

Buckets used to collect mortalities should have secure tight fitting lids that will exclude predators and scavengers. Buckets should be cleaned and disinfected before being returned to the fish rearing area.

After mortalities have been collected they should be stored (they may be frozen) in a central location away from the fish rearing area until they can be removed from the site.

Details of the Operating Procedure:

Mortalities are collected on a daily basis following feeding.

Morts are counted off of the screens during cleaning. They are also counted during vacuuming of the ponds. Morts vacuumed and swept off the screens are disposed of into the river via the fishway outflow.

Morts may be collected into perforated wash basins which are taken to the lab for counting and weighing.

If morts are from a different water system or numbers are excessively high, they are disposed of to the disposal holes on site.

Mortality collection equipment is dedicated for each stock and species being cultured. This equipment is not moved around the site, it is dedicated by area.

Forms & Records

References:
Outbreak Response

Outbreak – Disinfection Protocols
Mortality Collection and Disposal
Mortality Classification
Euthanasia
Equipment disinfection
Site and staff disinfection and biosecurity

4.21 Mortality Classification

Rationale: Mortalities must be examined for signs of disease to allow the early identification of developing problems. This SOP addresses section *2.3.3* and *2.3.3.1* of the General Principles of Fish Health Management.

The goal of this SOP is to ensure proper classification of dead fish into general categories to assist in the early identification of developing problems.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles and Details of the Operating Procedure:

All mortalities should be classified and recorded in the husbandry logs.

A certain degree of historical mortality is presumed but staff should record data and examine the mortality curve to observe any trends in mortalities over time. This is useful information as some hatcheries have disease signatures and these affect at different life stages etc but are specific to sites. If levels are seen to be above these then investigation should be initiated.

Standard mortality classifications may include:

Background Mortality (expected background losses)
Systems related (systems or equipment failure)
Environmental (water quality, plankton)
Fresh (fresh, unexpected apparently healthy losses or disease morts)
Handling/transport (losses related to handling or transport)
Matures (fish that have matured and died)
Old (morts are too old and decomposed to determine cause of death)
Predators (fish killed or injured by predators)
Culls/Quality Control/Poor Performers (fish intentionally removed from the population

Details of the Operating Procedure:

Mortalities are not formally classified. If mortalities increase and the cause is not obviously a feed issue or an environmental effect then a disease agent may be assumed issue and the Veterinarian will be contacted. Increases in mortalities will be noted in the comment sheets.

Forms & Records:
Mortality records
Enpro

References:

4.22 Outbreak Response

Rationale: Unexpectedly high losses may occur for any number of reasons, including a precipitous decline in water quality, environmental or feed-borne toxin, infectious disease, etc. **In the event of a fish health crisis or potential disease outbreak, until the cause of mortality has been confirmed, the site should be managed as though an infectious agent is present.** Steps must be taken to keep the pathogen load as low as possible and to prevent spread of the pathogen both within and from the site.

This SOP addresses section *2.3.4* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that the pathogen load is maintained as low as possible and spread of pathogens on or off the site is prevented.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

The procedures outlined in this SOP are set out as being appropriate by the DFO Veterinarian and are to be adhered to in the event of an infectious disease outbreak or a suspected disease outbreak

When an infectious disease problem is suspected the DFO Veterinarian should be ***immediately*** notified.

4.22.1 *Securing the Site*

At the Veterinarian's recommendation the site may be officially quarantined and this quarantine will remain in effect until such time as the problem has been diagnosed and/or managed. (See *Quarantine/Isolation Procedures for Suspected Disease Outbreaks*)

During an outbreak:

Visitors and non-essential staff should not be allowed on site unless previously authorized by the Fish Health Management Team

Hatchery management must notify other fish rearing facilities in the geographic area of the outbreak

Any suspected infected population should be quarantined/isolated from the healthy population, as are the items they may have contaminated (nets, buckets, siphons, etc).

Fish should not be handled any further and any movement of fish on/off and within the site should be halted.

The frequency of mortality collection should be increased during an outbreak but affected tanks should be mort picked last and staff should adhere to disinfection procedures between tanks and rearing units.

Where possible, separate equipment should be designated for the affected unit. All equipment, surfaces and clothing that come in contact with infected fish or infected material should be thoroughly disinfected after use to avoid potential transfer.

4.22.2 ***Assessment***

The Veterinarian must be sent all records and appropriate sampling information to determine cause of the outbreak and best course of action. The Veterinarian will provide instructions for proper sampling.

The Veterinarian will review management records including: species, age, year-class, source, vaccination, movements, treatments, results of previous diagnostic screening or disease events, water quality, feeding history, mortality rate for several weeks prior to the outbreak and fish behavior in the weeks previous to the outbreak.

Fish should be observed frequently and monitoring should continue after the initial workup to determine the course of the outbreak and to assess whether treatment and/or management measures are effective. Feeding response and water quality should be monitored and water and feed samples are to be taken if requested.

Healthy fish must be cared for first and personnel should disinfect themselves between handling groups to avoid inadvertent transfer.

Disinfection procedures (below) should be followed for movements into and out of the affected areas of the facility.

On site post mortems and sampling may be performed at the discretion of the Veterinarian and may be conducted by fish health personnel after securing the site. (See guidelines under *Diagnostic Sampling protocols*). Such samples must be properly handled, properly stored and promptly shipped (See *Sample Shipment to a Diagnostic Laboratory*) as per the Veterinarian's instructions to ensure that they will supply relevant information.

Temporal distribution of disease will most likely be assessed by biweekly sampling. Spatial distribution is assessed by conducting health checks on apparently healthy fish throughout the facility.

Further diagnostic testing to be conducted is at the discretion of the Veterinarian responsible for the case, which may include health checks on 60 randomly sampled fish and 20 moribund fish.

A treatment or action plan will be determined by the Veterinarian and hatchery management. The Veterinarian and site management will work together to review fish health records and the incident and make recommendations on how to avoid or handle similar events in the future.

4.22.3 ***Outbreak – Disinfection Protocols***

4.22.3.1 Personnel and Equipment

In the event of an outbreak, foot baths are to be used by all personnel before entering and leaving the facility. In the case of a diagnosed viral outbreak a 2% solution of Virkon™ is used in the foot bath. A 1% solution of Virkon™ should be used for dip net disinfection.

Foot baths should be clearly marked and a log of when the bath concentration has been tested or when it has been changed should be kept so all personnel are aware of its efficacy.

Raingear, field kits and boots of fish health personnel should be disinfected before entering and leaving the site.

There should be a separate disinfectant bucket and brush for fish health personnel visiting the site.

4.22.3.2 Mortalities

Mort collection equipment should be disinfected after use, especially during any incidence of pathogenic outbreak.

Dissection of fish for examination and/or samples must be conducted in a contained area to prevent the spread of disease within the facility.

Any surfaces in contact with dead fish are to be disinfected after contact.

Details of the Operating Procedure:

In the case of an unusual, unexplained or unexpected mortality rate the first step is to look for any obvious water problems.

When mortalities are greater than normal the rate of mort picking is increased, samples are forwarded to the Veterinarian and the pathology laboratory. Support Biologists will be contacted and informed of the problem.

If fish are not feeding, feed will be reduced or withheld and the pond will be quarantined as best as possible. Separation of people and gear will be exercised where possible. Buckets, nets and other equipment will be flagged with high visibility tape to ensure that all staff are aware that it may be contaminated and is restricted for use with the affected enclosure. Caution tape may be laid out to deter entrance.

A relatively common health issue at this site includes Myxobacterial infections, particularly in the coho. In these cases, Chloramine-T treatments will be undertaken. If this is observed to arise in one container,. All gear will be kept separated from that group.

Sampling plans or any other handling procedures that are planned are cancelled.

Every attempt to improve the environment is made. Flows are increased, feed is reduced, oxygen is injected (where available). Greater resources will be dedicated to sick groups to ensure that morts are picked frequently and no dead fish are remaining in the enclosure.

If the issue becomes chronic and attempts have been made to improve their condition for several weeks, they may be moved into salt water which has been found to assist in recuperation. Under these circumstances the fish will be moved to the lake pens. If this seems unlikely to assist the fish, they may be released.

Forms & Records

Quarantine Notice

References:

Quarantine/Isolation Procedures for Suspected Disease Outbreaks
Diagnostic Sampling protocols
Sample Shipment to a Diagnostic Laboratory
http://www.syndel.com/d_p_f_s/Virkon™_info_sheet.html
http://www.syndel.com/d_p_f_s/dilution_testing_kits.htm
http://www.syndel.com/msds/Virkon™_msds.html

4.23 Quarantine/Isolation Procedures for Suspected Disease Outbreaks

Rationale: Quarantine is the enforced physical separation of the healthy population from a (potentially) infected population, their products or items they may have contaminated. [1] This will prevent transmission within and between facilities. It is virtually impossible to completely treat all effluent from our facilities, therefore this is not "truly" a quarantine as we cannot effectively halt all spread from an infected population in our facilities. However we can isolate our fish from pathogens entering the site with due diligence. This SOP addresses section *2.3.4.3* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that disease transmission within and between facilities is prevented during a disease outbreak.

The procedures outlined in this SOP are set out as being appropriate by the DFO Veterinarian and are to be adhered to in the event of an infectious disease outbreak or a suspected disease outbreak

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly

General Principles:

In the event of a diagnosed pathogenic outbreak:

4.23.1 ***Securing the Site***

Affected hatcheries should be quarantined/isolated; facilities locked down. Gates to the facility should be closed and only essential personnel admitted.

Disinfection procedures should be followed for movements into and out of the facility.

The movement of fish, vehicles, equipment and personnel from the affected hatchery to fish bearing habitat or other fish rearing facilities should be immediately halted.

4.23.2 ***Isolation of Infected Group***

The affected fish rearing containers should be isolated, movement of fish into and out of these containers stopped.

If possible, effluent is trapped and treated prior to discharge to the environment.

4.23.3 ***Mortality Removal***

Depending on overall morbidity rate, all sick, slow swimming or moribund fish should be removed from the environment. Mortality removal should be done at least twice daily.

[1] Martin et. al., eds. Veterinary Epidemiology: Principles and Methods.

Morts should be collected into spill proof containers with secure lids and transported to a composting landfill for disposal. Equipment and containers used to collect mortalities should be disinfected after each use.

Forms & Records

References:

Outbreak Response

Outbreak – Disinfection Protocols

4.24 Juvenile Treatments

Rationale: Due to a developing immune system and the physiological stress related to growth and smoltification, juveniles represent a particularly susceptible life stage. Judicious use of antimicrobial agents may help minimize losses due to infectious agents. This SOP addresses section *2.1.3.10* of the General Principles of Fish Health Management.

Often, the combination of historical disease incidence combined with clinical signs, can allow a presumptive diagnosis of a disease agent by hatchery staff. The commonly used external antimicrobial agents listed herein do not require a veterinary prescription. However, diagnostic sample submission and consultation with the Veterinarian is encouraged before attempting any treatment, and is strongly recommended in the event of treatment failure to produce the anticipated improvement in levels of morbidity and/or mortality. The goal of this SOP is to ensure safe administration of externally applied antimicrobial agents to minimize loss of juveniles during rearing.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Fish should be held off food for 24 – 72 hours prior to treatment. This serves to reduce fecal fouling of the tank and lower the metabolic demands of the fish.

Tanks should be carefully siphoned/cleaned to remove as much detritus as possible before treatment. External antimicrobial agents will act on whatever organic matter is present; cleaning the tank helps ensure the highest activity against the pathogen in question. A high level of organic matter will render the treatment ineffective.

It is always safest to treat a small 'test' group of fish initially to detect errors in calculations during solution preparation and administration or any unusual species/stock/strain sensitivities to the treatment chemical.

Fish should be monitored closely during treatment for any adverse effects. Gathering at the inflow, gasping at the surface, attempts to jump out of the water, etc. should be considered signs of treatment toxicity. The treatment should be stopped ASAP and the water flow increased to rapidly dilute and flush out the offending chemical. The Fish Health Management Team should be informed and the details of the procedure (including tank volume, flow, concentration calculations, chemical expiration and storage, stock and working solution preparation, etc.) should be reviewed. Fish should continue to be held off food for a 24 – 48 hour recovery period before attempting further treatments.

Details of the Operating Procedure:

4.24.1 ***Chloramine-T***

Chloramine-T is a disinfectant used for surface bacterial infections including bacterial gill disease and fin rot. Chloramine-T powder can cause burns or sensitization on skin contact and sensitivities upon inhalation; it is injurious to eyes and is harmful if swallowed. Staff should review WHMIS information prior to handling this product and employ appropriate personal protective equipment.

Chloramine-T is a gill irritant to fish; its toxicity is increased by soft water and low water pH. It is highly advisable to determine your water pH and hardness prior to using Chloramine-T to protect the fish. Where water chemistry is unknown, the low end of the recommended dosing range should be used to minimize the chances of toxic effects on fish.

The standard flush, bath or dip treatment with Chloramine-T is 8.5 – 12 ppm for one hour daily on three treatments on consecutive or alternating days.

Chloramine T will be diluted to 2 mg/L and discharged. If more concentrated solutions are to be discharged they will first be treated with Sodium thiosulphate to minimize environmental impact. At Nitinat there is generally a 20 – 50 fold dilution when this compound is being used.

At the first sign of infection consult with Veterinarian, and submit samples to the Fish Pathology at PBS for diagnosis.

4.24.2 ***Calculate Chloramine T Required***

To determine the amount of Chloramine-T required:

1. Multiply flow by 60 to determine liters per hour (L/hr)

2. Multiply L/hr by x mg/L of flow (mg as instructed by the Veterinarian)

   ie: flow 1000 L/min x 60 = 60,000 L/hr

   60,000 L/hr x 12mg/L = 720,000 mg

   720,000/1000 = 720g of Chloramine-T per treatment

3. Then multiply the amount of treatment chemical required by the number of containers that need to be treated and the number of days the treatment is prescribed. This will give you the amount of Chloramine-T required

4. Weigh out Chloramine-T into daily treatments for each container

Read MSDS sheet before proceeding

4.24.3 *Preparation*

Equipment Required:

- □ Calculator
- □ Scale
- □ White "Kitchen catcher" garbage bags
- □ Scoop
- □ Permanent marker
- □ Chloramine T
- □ Gloves
- □ Dust mask
- □ Eye protection
- □ 6 wheel cart
- □ Clean garbage bucket
- □ Varistaltic pump
- □ Extension cord
- □ Ground fault relay
- □ Stopwatch
- □ 500ml graduated cylinder
- □ Brick
- □ Stir stick
- □ Eye protection

1. Weigh daily treatments into Kitchen catchers and write the container # on side of bag.
2. Clean up any "spillage" around scale.
3. Switch infected container to ground water
4. Vacuum container before treatment
5. Mix Chloramine-T
6. Treatment is for 1 hour in duration
7. Mix daily prescription into 24 L of warm water
8. Mix well
9. Place Varistaltic pump on cart
10. Place bucket of Chloramine-T solution on cart
11. Put intake tube into bucket
12. Carefully move cart to container to be treated
13. Plug in Varistaltic pump to power using ground fault relay
14. Place brick under one edge of bucket to tilt to one side to aid in pumping out all of treatment
15. Turn on Varistaltic pump
16. Allow pump to run until a consistent amount of treatment is discharging
17. Using stopwatch adjust speed of pump until it will fill the graduated cylinder to 400 ml in 60 seconds. This speed will deliver treatment in 1 hour. (24liters /400ml = 60)
18. Place discharge end into inflow of container to mix
19. Cover pump to keep rain off
20. Monitor to assure delivery speed

21. Upon completion of treatment remove the delivery hose from the container and place it in a pail of warm water
22. Rinse out bucket
23. Put away all equipment

4.24.3.1 MSDS information

Chloramine-T powder can cause burns or sensitization on skin contact and sensitivities upon inhalation; it is injurious to eyes and is harmful if swallowed. Staff should review WHMIS information prior to handling this product and employ appropriate personal protective equipment.

4.24.4 ***Parasite-S™ (Formalin)***

Parasite-S™ is the trade name for an anti parasitic, formalin-based solution that is commonly used against parasitic and bacterial gill disease including those caused by the protozoans *Ichthyobodo* (Costia) and *Trichodina*. It is also a standard disinfectant used in hatcheries for the prevention and treatment of egg fungal infections (see *Egg Fungal Treatments*).

The normal dilution of formalin for treating fish is 1:6000 or 167 ppm. This concentration is achieved by combining 17 mls of Parasite-S per 100 litres of water. Exposure is normally 30 – 60 minutes daily, and may be done on consecutive or alternating days for three treatments in total.

Cautions:

Formalin should never be added to water containing fish without first diluting it and then mixing it in thoroughly to avoid 'hot spots'.

Formalin should not be used if :

- Dissolved oxygen of the water is <5ppm
- The water temperature is >27^{o}C
- Heavy phytoplankton growth is present.

4.24.4.1 Parasite-S MSDS information:

Formalin must be handled with care. It is harmful if inhaled, and can seriously irritate eyes and skin after contact. Formalin should only be handled in well ventilated areas, preferably while wearing a respirator and safety glasses. Staff should review WHMIS information prior to handling this product and employ appropriate personal protective equipment.

Parasite-S contains methanol which inhibits the formation of paraformaldehyde, a white precipitate that is extremely toxic to fish. Even so, formalin should be stored in the dark above 10^{o}C and not allowed to freeze. If Parasite-S is allowed to freeze, it should be discarded due to the rapid formation of paraformaldehyde.

The toxicity of formalin increases as temperature increases. In very soft water, concentrations of formalin should not exceed 25 ppm. Concentrations greater than 250 ppm may cause severe gill damage and should not be used on salmonids.

Formalin is a gill irritant and thereby reduces gas exchange. This is especially of concern as formalin is commonly used when fish gill function may already be compromised. Additionally, formalin is a reducing agent that absorbs oxygen from the water. Therefore, the safest course is to treat with formalin at the time of day when the water temperature is at its lowest, and to provide supplemental oxygenation via airstones. The fish should be closely monitored during treatment for signs of respiratory distress (increased opercular movements or gasping at the surface) and the treatment terminated if needed.

Forms & Records:

References:

Egg Fungal Treatments
http://www.syndel.com/d_p_f_s/parasite-s_info_sheet.html
http://www.syndel.com/handling/transportation_of_live_fish.html
http://www.syndel.com/handling/vidalife_info_sheet.html

4.25 Broodstock Treatments

Rationale: Broodstock are a sensitive life stage. They are channelling their energy stores into the maturation of their gametes, and undergoing the physical stresses related to migration, changing temperatures and re-entry into freshwater. The cumulative effects of these multiple stressors can result in fish whose immune system is compromised. As a result, broodfish may be shedding pathogens in increased numbers and may have an increased susceptibility to secondary, opportunistic infections. Broodstock treatments can help reduce pre-spawning mortality losses and can help reduce the risk of vertically transmitted pathogens being passed to the offspring. Brood fish can also be treated to synchronize spawning dates. This SOP addresses section *2.2.9* of the General Principles of Fish Health Management. The goal of this SOP is to ensure successful treatment of broodstock to lessen pre-spawning mortalities, reduce vertical transmission of pathogens in areas with a historically high prevalence of an antibiotic-susceptible pathogen and/or to synchronize spawning.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Fungal infections

Brood fish are particularly susceptible to external fungal infections which can contribute to pre-spawning mortalities. Routine use of mucus protectants (e.g. Vidalife™) during handling events for ripeness checks and gender segregation, can help reduce the incidence fungal infections. Once established, antifungal formalin baths or salt treatments can be used to reduce the severity of fungal infection.

Bacterial infections

Based on historical disease patterns and/or pre-spawning mortality rates, antibiotic therapy may be indicated to reduce brood losses and as a prophylactic treatment to help decrease the vertical transmission of bacterial pathogens, such as *Renibacterium salmoninarum*, the causative agent of Bacterial Kidney Disease. Decisions to pursue antibiotic therapy must be made in consultation with the Fish Health Veterinarian. Antibiotics must be obtained and used as directed by veterinary prescription.

Broodstock antibiotic injections typically are given one month before the anticipated spawning date, as peak concentrations of antibiotic are present in the gametes 2 and 4 weeks post-injection. For the reduction of vertical transmission of bacteria, it is reasonable to inject the females only, however, if pre-spawning mortalities are high, treating both males and females may be beneficial.

Spawning synchronization

Manipulating environmental cues such as temperature, photoperiod, nutrition, holding density, etc., is critical for the maintenance of natural reproduction rhythms. Captive brood programs experience unique challenges in terms of the synchronization of spawning readiness. Under veterinary

prescription and supervision, hormonal injections (e.g. Ovaplant™, Ovaprim™) may be given to broodfish to help synchronize the timing of spawning.

Details of the Operating Procedure:

4.25.1 *Parasite-S™ (Formalin)*

Parasite-S™ is the trade name for an anti parasitic, formalin-based solution that is commonly used against parasitic, fungal and bacterial gill infections. It is also a standard disinfectant used in hatcheries for the prevention and treatment of egg fungal infections (see *Egg Fungal Treatments*).

The normal dilution of formalin for treating fish is 1:6000 or 167 ppm. This concentration is achieved by combining 17 mls of Parasite-S per 100 litres of water. Exposure is normally 30 – 60 minutes daily, and may be done on consecutive or alternating days for three treatments in total.

Cautions:

Formalin should never be added to water containing fish without first diluting it and then mixing it in thoroughly to avoid 'hot spots'.

Formalin should not be used if :

- Dissolved oxygen of the water is <5ppm
- The water temperature is >27^{o}C
- Heavy phytoplankton growth is present.

Adults may be treated with Parasite-S using the peristaltic pump

1. *Contact the veterinarian regarding treatment and dose*
2. *Starve fish for 24 hours*
3. *Check the dissolved oxygen concentration in the enclosure prior to treatment and during treatment*
4. *Clean the tanks of organic matter to reduce waste of treatment compound*
5. *Make up stock solution of Parasite-S (can you send me a sample calculation that you use and how it is mixed up)*
6. *Set up peristaltic pump and hoses and buckets (how?)*
7. *Calibrate the pump according to the manual instructions (Manual is kept where?)*
    a. *Dispense X volume of water into a graduated over a period of one minute*
    b. *If a volume of X does not dispense, or if more dispenses in this time period, recalibrate the pump*
    c. *Treatment volume is what?? of stock solution dispensed at a rate of what? over how long?*
8. *Inform other staff which fish are being treated so they are not fed or disturbed during treatment*
9. *Reduce flows if necessary yes or no? To what if yes?*

10. *Place treatment dispensing hose into header of the container to be treated and turn pump on*
    a. *Monitor during treatment to ensure that distress is not evident. If the fish behaviour changes– frantic behaviour, avoidance, etc (….what…what do you do…?)*
11. *When the treatment is completed, return flows to normal if they were reduced*
12. *Flush and clean all equipment and put away in its appropriate location*
13. *Follow up with veterinarian*
14. *Withhold feed until the following day*

4.25.1.1 Parasite-S MSDS information:

Formalin must be handled with care. It is harmful if inhaled, and can seriously irritate eyes and skin after contact. Formalin should only be handled in well ventilated areas, preferably while wearing a respirator and safety glasses. Staff should review WHMIS information prior to handling this product and employ appropriate personal protective equipment.

Parasite-S contains methanol which inhibits the formation of paraformaldehyde, a white precipitate that is extremely toxic to fish. Even so, formalin should be stored in the dark above 10^{o}C and not allowed to freeze. If Parasite-S is allowed to freeze, it should be discarded due to the rapid formation of paraformaldehyde.

The toxicity of formalin increases as temperature increases. In very soft water, concentrations of formalin should not exceed 25 ppm. Concentrations greater than 250 ppm may cause severe gill damage and should not be used on salmonids.

Formalin is a gill irritant and thereby reduces gas exchange. This is especially of concern as formalin is commonly used when fish gill function may already be compromised. Additionally, formalin is a reducing agent that absorbs oxygen from the water. Therefore, the safest course is to treat with formalin at the time of day when the water temperature is at its lowest, and to provide supplemental oxygenation via airstones. The fish should be closely monitored during treatment for signs of respiratory distress (increased opercular movements or gasping at the surface) and the treatment terminated if needed.

Forms & Records:

References:

Egg Fungal Treatments
Anaesthesia
Fish Handling Procedures

4.26 Top-Coating Medicated Feed

Rationale: Small volumes of medicated feed are required at sites with lower production numbers. At these sites, medication may be top-coated unto appropriate food and fed as required by veterinary prescription. The goal of this SOP is to ensure that medicated feed is mixed properly.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

All staff members who will be handling medicated feed are required to be familiar with the MSDS information and take precautions to protect their health.

General Procedures:

Feed should be obtained from a feed mill that has been inspected by the CFIA.

Medicated feed should be clearly identified in the storage area by use of different colored bags and clear labels. It should be physically separate from non medicated feed. Medicated feed should be fed out according to veterinary prescription. All equipment which comes in contact with medicated feed should be cleaned and disinfected thoroughly after use.

Details of Procedure:

Always use PERSONAL PROTECTIVE EQUIPMENT and avoid exposure. A properly fitted respirator must be worn when dispensing antibiotic powder. Eye protection, gloves and protective clothing must also be worn during top coating of feed.

1. *Contact the Veterinarian for the prescription and dose*
2. *Determine daily feed requirements for fish being treated*
3. *Fish will be fed at half ration during treatment to encourage feed uptake*
4. *Measure out 10 days worth of feed based on the above calculations*
5. *Mix in some of the next smaller feed size to ensure that smaller fish receive feed and treatment*
6. *Don personal protective equipment (gloves, respirator eye protection etc)*
7. *Measure out the appropriate amount of drug for the feed to be delivered (contact veterinarian for dosage)*
8. *Measure out approximately 1 L of warm water and add Knox gelatine powder. Allow to dissolve*
9. *Pour gelatine solution into a spray bottle*
10. *Place feed into cement mixer*
11. *Place drug into the cement mixer with the feed*

12. *Turn on cement mixer and allow the drug to distribute in the feed*
13. *Lightly mist the feed with the gelatine solution. Do not apply too heavy a coating of solution.*
14. *Continue to run cement mixer until feed appears evenly coated*
15. *Distribute medicated feed into the number of labelled, lidded feed buckets required for the duration of the medication period*
16. *Place medicated feed in the cooler away from other feed to ensure confusion does not occur*
17. *Clean equipment and medicated feed containers following treatment. To clean the mixer, add water and one or two shovels of gravel to the mixer. Allow the mixer to run and polish the sediment off the insides. Rinse well.*
18. *Clean buckets with soap and water and disinfect them after they have been emptied*

Medicated fish must not be available for human consumption (or released) for 90-120 days following the final treatment day. Consult the Veterinarian for further information.

Forms & Records


DRUGS.XLS


Top Coating Food STEPS

References:

4.27 Medicated Feed: Storage, Handling, and Feeding

Rationale: This SOP addresses section *2.3.3.3* of the General Principles of Fish Health Management.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Medicated feeds must only be used under veterinary prescription with accompanying caution for withdrawal times. Rational antibiotic selection will be based on the Veterinarian's clinical judgment, the use of antimicrobial sensitivity testing, and due diligence with regards to the prevention of promoting antibiotic resistant bacteria.

Medicated feed should be clearly identified. Daily rations may be kept in seal-top container at tank side, with remaining feed refrigerated.

Persons dispensing medicated feed should wear protective latex gloves.

Medicated feed prescription amounts are based on the most accurate information available regarding the size and number of fish to be treated. Ensure that appropriate information has been forwarded to the veterinarian to ensure proper prescription. All medicated feed should be fed out as directed for the total number of days prescribed, leaving no food leftover.

Failure to see expected improvements after 5 days on medicated feed should be reported to management and the prescribing Veterinarian.

Adverse reactions to medicated feed should result in immediate cessation of the treatment and consultation between Fish Health Management and the prescribing Veterinarian.

During a treatment course with medicated feed no handling (i.e. sampling, marking, grading etc.) should be carried out. ***Once medicated feeding is initiated, it must not be discontinued without the approval of the prescribing Veterinarian.***

Medicated feed is not as palatable and the fish may refuse to eat it. It is important to closely monitor the feeding response of the fish when starting treatment to ensure:

- That fish are consuming the food without subsequent regurgitation
- The feeding rate is slow enough to prevent excess feed from reaching the tank bottom
- There is adequate coverage to all areas of the tank/raceway

Staff may help encourage eating by holding fish off food for 24 - 48 hours before starting medicated feed or by initially mixing decreasing amounts of non-medicated food for the first couple of feedings. Palatability can also be increased by the addition of krill or garlic powder. This may be especially valuable at temperatures < 5^{o}C.

If a feeding hierarchy exists within the tanks, it may be safest to start each day feeding a few handfuls of non-medicated feed to take the edge off the more aggressive feeders to ensure they don't consume a higher proportion of medicated feed than other fish. This should be followed with the daily allotment of medicated feed given at normal feeding intervals. After the daily medicated feed ration has been fed, the fish may be fed to satiation with non-medicated feed.

At high water temperatures, lower oxygen solubility can become a limiting factor for fish culture. For health compromised fish, this is even a greater concern as any stressor can increase the risk of death. If temperatures are >16^{o}C, dissolved oxygen levels should be measured 2 hours after feeding to ensure adequate dissolved oxygen is present. If dissolved oxygen measures < 6.0 mg/L, the timing of feeding should be changed to avoid mid-day, when water temperatures are highest.

Details of the Operating Procedure:

Wear all personal protective equipment when handling medicated feed.

Wash all equipment thoroughly after handling

No palatability additives are used in medicated feed.

Feed fish at half regular ration levels to encourage feed uptake.

Mix one size smaller feed with regular feed size to ensure that smaller fish in the container access the medication.

Feed fish the medicated portion of their daily ration as the first 25% of the daily half-ration feed fed out. Feed non-medicated feed for the remainder of their half-ration.

Forms & Records:

References:

4.28 Diagnostic Sampling protocols

Rationale: Samples of fish for diagnostic purposes must be collected properly to ensure that results obtained are useful. This SOP addresses section *2.3.3* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that samples are collected properly.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

The best fish to sample are moribund fish. Morts should only be sampled if their gill tissue is red.

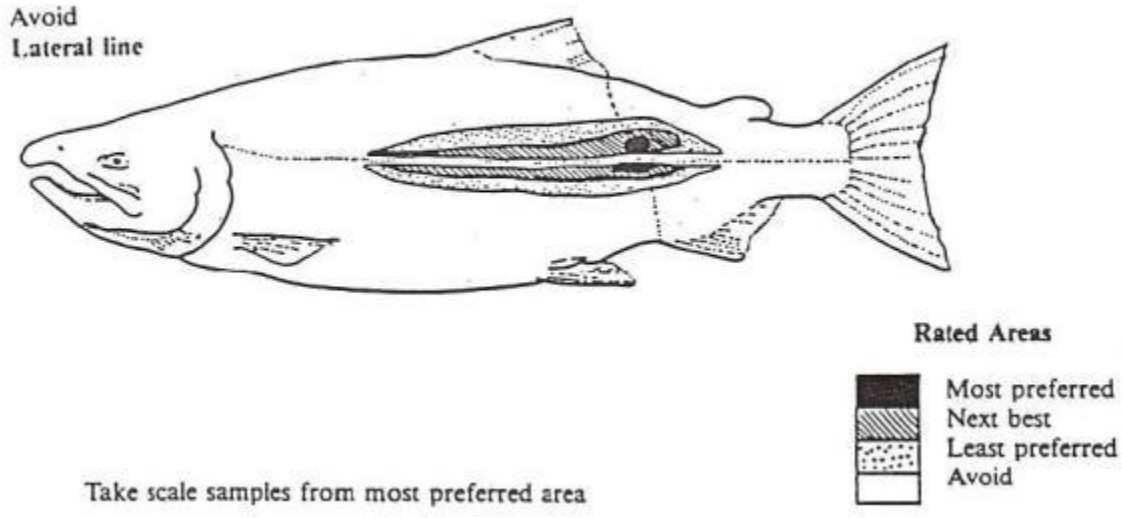
Samples may be collected for parasitology, bacteriology, virology or histology.

Fish must be humanely euthanized prior to sample collection. It is preferable to do this by an over dosage of anaesthetic as blunt trauma can cause gill aneurysms and make interpretation of some histopathology difficult. (see *Euthanasia*).

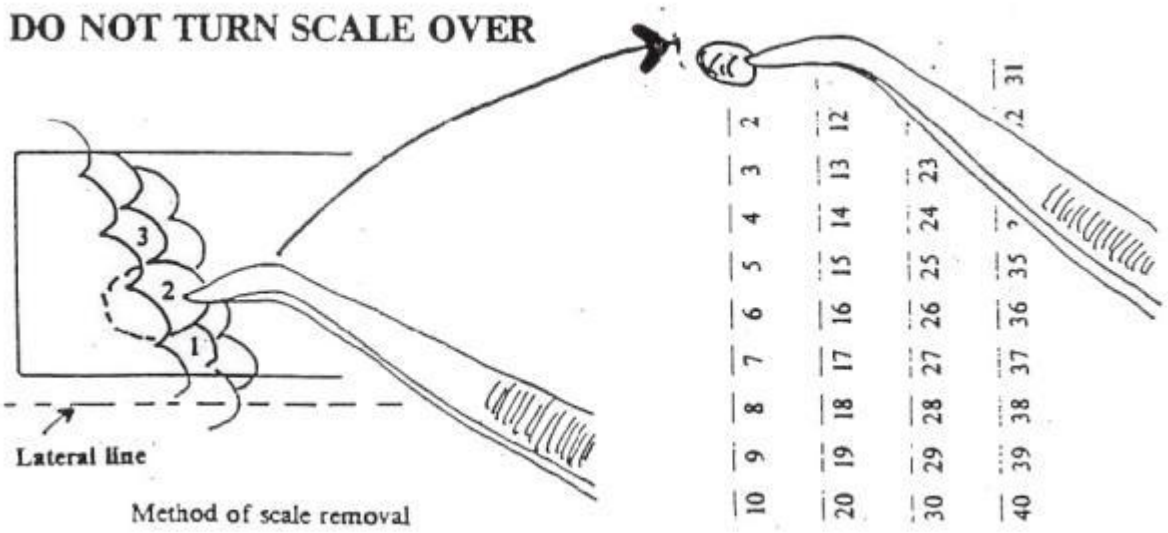
Bleeding fish prior to sampling can be done to limit the amount of blood pooling during sample collection; the gill arch can be cut on large fish and the tail cut off on smaller fish.

Details of the Operating Procedure:

4.28.1 ***Scale Sampling***



1. Place the fish on its side.
2. Wipe the preferred area clear of water and mucus.
3. Remove the scale by grasping its exposed edge with tweezers arid pulling.
4. Hold the scale up to a light source to check for deformation or regeneration. Scales with these signs are unreadable. Take another sample scale from the next rated area. For further explanation please contact the morphology lab.
5. Center the scale on the numbered square on the scale book so that the side that faced up on the fish remains facing up on the book. (See Figure below.)
6. To check if the correct side is facing up, scrape the surface of the scale. It should feel rough with ridges.
7. Be sure the scale is sticking firmly to the scale booklet.
8. Turn the fish on its other side and repeat the procedure.
9. The scale book surfaces *must* be kept dry. Excessive moisture will dissolve the book's adhesive coating and cover the scale or wash away the adhesive so that the scale cannot stick to the book.



Method of scale removal

4.28.2 ***Tissue sampling for BKD diagnositcs***

Follow the procedures outlined in the Appendix (BKD sampling procedure (revised 2006))

4.28.3 ***Otolith Sampling***

4.28.4 ***Otolith Sampling***

Equipment

- Sharp, thin knife
- Forceps or tweezers
- Numbered containers (small plastic tubes are preferred)
- Data sheets

1. Locate the cranial otolith pockets by cutting directly down from the top of the head, approximately midway, between the eye and the operculum
2. Collect both otoliths, unbroken and as clean as possible with forceps or tweezers
3. Place otoliths in numbered containers, usually small plastic tubes
4. Record all information that is needed for each individual fish sampled. (Date, area, species, sex, fin clip, length)
5. Once all otolith are taken for the sampling season, send samples by courier to the lab for analysis

Forms:


Example adult sampling plan


Biosampling Form

References:

Euthanasia

4.29 Sample Shipment to a Diagnostic Laboratory

Rationale: In order for accurate diagnoses to be obtained, fish samples must arrive at the laboratory in suitable condition. The goal of this SOP is to ensure correct transport of live or dead fish to the fish pathology laboratory in order to receive valid results. The goal of this SOP is to ensure that fish samples are shipped in a manner that protects the sample integrity.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Deciding when diagnostic support is needed can often be difficult. Shipping samples is time consuming and costly. If mortalities are unexpected, clinical signs are suggestive of a disease of concern (eg. popeye and/or swollen abdomens at a facility with a history of recurrent BKD infection), or if the daily mortality rate exceeds 0.5% of the population, a sample of the affected fish should be transported to the Fish Pathology Laboratory at the Pacific Biological Station.

Equipment List:

- □ Shipping container
- □ Elastic bands
- □ Packing tape
- □ Ice or freezer packs
- □ Ziplock bags
- □ Disinfectant
- □ Heavy duty plastic bags
- □ Oxygen supply
- □ Submission form
- □ Newspaper
- □ Waterproof labels
- □ Waterproof marker

Details of the Procedure:

4.29.1 ***Before shipping:***

Collect fish history information, including: population size, clinical signs, mortality and morbidity rate, diet and feed consumption, water quality conditions, records of recent stressful events (e.g. low water event, marking), vaccination status, disease and treatment history.

Contact the fish pathology laboratory technical staff at the Pacific Biological Station, Rm T308, 3190 Hammond Bay Road, Nanaimo BC, V9T 6N7. Phone (250) 756-7057; Fax (250) 756-7053, westbyc@pac.dfo-mpo.gc.ca

or

Contact the DFO staff Veterinarian at the PBS, Dr Christine MacWilliams at (250) 729-8377, macwilliamsc@pac.dfo-mpo.gc.ca

A determination will be made whether live or dead fish are required for evaluation. In most cases live fish are preferable for diagnostics; however, for some situations dead fish may suffice.

Ensure the diagnostic lab is aware of fish on their way to the lab and provide the estimated time of arrival.

Fish must arrive at the diagnostic laboratory by mid day at the latest to ensure staff has time to work on the fish.

4.29.2 ***Selecting the samples:***

Where possible, select moribund fish for shipment. Seek advice from the Veterinarian and fish health management to determine how many fish and from which locations the fish should come.

Freshly captured, live fish that display signs of the problem are the ideal samples to collect for submission. The diagnostic lab may also request a sample of apparently healthy fish from the population.

There may be a need to randomly sample apparently healthy fish from the population; rely on veterinary advice for this decision.

4.29.3 ***Shipping live fish:***

Precede the shipment by a call or an email to make the lab aware that samples are to be forwarded and request any pertinent information or guidance.

Wrap ice packs in newspaper and place into the bottom of a hard sided box or cooler that is to be used for shipping. Alternatively, ice may be double bagged in sealed zip-lock bags and placed in the bottom of the container. Newspaper should be placed on top of the bags of ice.

Fill a silver wine bladder 1/3 full with well water. Using a small funnel, place approximately 30-35 fry into the wine bladder.

Fill the remaining 2/3 of the volume of the bladder with oxygen and secure the snap lid.

Separate moribund and apparently healthy fish

Place the wine bladder(s) into the cooler.

Include a Sample submission form describing the fish population and the problem and/or an accompanying letter with more detail.

Secure the container with duct tape to prevent accidental spillage. Spray or wipe down the outside of the container with an appropriate surface disinfectant.

Transport the samples to the Pacific Biological Station or contact a courier for overnight delivery. If using an overnight courier, ship in the late afternoon so the fish are not in transit for as long. Ship fish

on Thursday afternoon at the latest to ensure prompt delivery to the Pathology lab. It is preferable to shiop samples early in the week.

4.29.4 ***Shipping fresh dead fish:***

In the event that no moribund fish are available for sampling, morts can be shipped. **Evaluate fish condition prior to shipping.** Only ship morts which still have red gills otherwise it is doubtful that any useful information can be gained from the samples.

If fish are too large to realistically send alive, freshly euthanized fish may be sent for diagnostics.

Fresh morts (red gills, firm flesh) should be placed in labeled, sealed double plastic bags without water. Ship dead fish in a container on ice as described above for live fish. Fish should not come in contact with the ice or freezer packs.

4.29.5 ***Shipping samples that have been collected from the fish:***

Place tissue samples in clearly labelled Whirl-pak bags

Place Whirl-pak bags into ziplock baggies

Tissue samples must be maintained at 4-7°C for shipment and they must be reached by the diagnostic laboratory within 24 hrs of sampling. Therefore it is best to ship the samples on the day that they are taken.

Place ice packs into the bottom of a small sandwich cooler. Place a layer of newspaper or paper towelling on top of the ice packs.

Place the double bagged samples in the cooler and seal with duct tape

Transport the samples to the diagnostic laboratory or ship via courier. If samples are shipped late in the afternoon via overnight courier they should arrive at the laboratory by noon the following day.

Note: Fish being forwarded to PBS for otolith analysis are *frozen individually and placed into ziplocks. The ziplock bags are placed into a cooler containing ice packs. The number of fish being forwarded will be dictated by the laboratory. Samples are generally held until a staff member is traveling to PBS*

4.29.6 ***Following Transport:***

Follow up with diagnostics laboratory to confirm receipt of the samples and tentative time frame for diagnosis and treatment recommendations.

Note: if the Fish Pathology Lab is unable to accept your samples due to scheduling conflicts, alternate user-pay diagnostic facilities are available:

Animal Health Centre	***Microtek International, Inc.***
B.C. Ministry of Agriculture and Lands.	*6761 Kirkpatrick Crescent*

1767 Angus Campbell Road
Abbotsford, British Columbia
V3G 2M3
contact: Dr. Gary Marty
phone 604-550-3003
fax 604-556-3010

Saanichton, British Columbia
V8M 1Z8
contact: Tim Hewison
phone: 1-800-667-5062 (ext. 201)
fax: 250-652-4802

Forms & Records

Sample submission form (see Appendix)

References:

Diagnostic Sampling protocols

Chemicals & Disinfectants

Veterinary Chemicals for use in Fish/Egg Disinfection in Canadian Fish Hatchery and Aquaculture Facilities

Fisheries and Oceans Canada (DFO) as the lead federal department for aquaculture management for Canada is providing the following list of chemicals that can be administered under the regulatory authority of Health Canada's Veterinary Drug Directorate (HC-VDD) for fish egg disinfection in facilities producing fish for food directly (via aquaculture) and indirectly (via salmonid enhancement programs). The Canadian Food Inspection Agency (CFIA) is responsible for monitoring to ensure that fish and fish products meet the requirements of the Fish Inspection Act and the Food and Drugs Act, and will take appropriate regulatory action when unapproved or banned substances are found. Since 1992, a prescription from a licensed veterinarian is required for the use of many veterinary drugs in hatchery and aquaculture facilities.

The following list and comments related to use for fish egg disinfection has been provided by licensed fish health veterinarians and complies with the regulatory requirements of Health Canada and the CFIA. A licensed veterinarian should be consulted to determine the appropriate treatment and dosage that is required for any of these treatments.

Approved Veterinary Chemicals

Note – the use of any other chemical for egg disinfection of food fish is illegal

Parasite-S® or Formalin-R Solution is a formalin-based solution that also contains methanol to prevent the formation of paraformaldehyde, which is toxic to fish. It is used as a bath to control external parasites on the gills, skin and fins of fish and to surface disinfect eggs.

Perox-Aid™ (hydrogen peroxide) is an antifungal agent for use on fish eggs. In Canada, it is the only product that is officially approved for use on food fish.

Salt Solutions. High salt concentrations can kill surface parasites and fungal infections. Treated fish may produce extra mucus from the skin and gills which assists in the removal of external parasites. Care must be taken with the concentration of salt used to ensure that treated fish do not experience osmotic shock.

Veterinary Chemicals Available through Emergency Drug Release (EDR)

Pyceze® (Bronopol) is an antimicrobial agent that is used as a preservative in health care, food-contact materials and cosmetics as well as an antifungal treatment for salmonid eggs. It is not approved for use in fish hatcheries in Canada and is only available by prescription from a licensed veterinarian through Health Canada's Emergency Drug Release program. Use of this drug should be discussed with your provincial or private fish health veterinarian

Ovadine® is under review by the VDD for approval as a fish egg disinfectant. Until approval is received, it is available only by prescription from a licensed veterinarian, through Health Canada's Emergency Drug Release program.

For more information, contact:
National Registry of Aquatic Animal Health
Fisheries and Oceans Canada
200 Kent St
Ottawa, ON K1A 0E9
Nrfd@dfo-mpo.gc.ca

4.30 Anaesthesia

Rationale: All operators need to anaesthetize fish from time to time for handling, vaccinating, marking or sampling procedures. Anaesthesia is used to minimize the stress of such procedures. This SOP addresses section *2.1.3.1* of the General Principles of Fish Health Management.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Prior to Anaesthesia:

Approved anaesthetics are obtained by veterinary prescription. ***The only anaesthetic agents registered for veterinary use with fish in Canada are TMS and Aquacalm.***

Health risks associated with handling the fish should be reviewed by fish health staff and the Veterinarian prior to procedure. All staff handling anaesthetics must be aware of WHMIS information.

Fish should be taken off feed for 24 to 72 hrs prior to being anaesthetized. Determination of the time of fasting includes consideration of fish size, diet, water temperature and existing knowledge about gut emptying times.

Anaesthetic baths should be prepared according to manufacturer's directions and should be made up using water from the enclosure the fish are being taken from to minimize stress.

During Anaesthesia:

Staff should wear personal protective equipment to minimize exposure to anaesthetic agents. Many agents are known to present health risks, others are currently unknown. It is advisable to be pro-active in preventing personal risk. Recommended gear includes safety/splash glasses, dust mask, latex or nitrile gloves and rubber boots.

Fish should be handled gently using nets with smooth surfaces. Larger fish should be supported ventrally when handled by hand; smaller fish should be handled with a dip net. Any dropped fish or jumpers should be handled by net instead of hands.

Mucus protectants (e.g. Vidalife™) may be employed to minimize damage to the fish mucus-skin barrier (see the *Fish Handling Procedures*).

A few fish should be tested first before adding larger numbers of fish to the anaesthetic bath, as the effect of an anaesthetic may vary with local water conditions, as well as the species, life stage and size of the fish. This step ensures that an incorrect dose will result in minimal losses.

The action of anaesthetics is affected by water quality. Water quality parameters, especially temperature and dissolved oxygen, should be monitored during the procedure. The temperature of the rearing unit and the anaesthetic and recovery baths should not differ by more than two degrees.

Visual behaviour of fish should be monitored during the anaesthetic bath. Fish should be observed for signs of visible distress or cessation of opercular activity which can be life threatening.

Airstones should be placed in the anaesthetic solution, with the airflow regulated for small bubbles to optimize oxygen exchange.

Fish should never be left unattended while in the anaesthetic bath. Once the desired plane of anaesthesia is reached, the fish may be removed and the handling/procedures performed.

More fish may be transferred to the anaesthetic bath as required but any degradation in water quality or change in the time to anaesthesia should be monitored and addressed. When induction time increases to > 2 minutes or water quality deteriorates (D.O. < 5 mg/L and/or temperature changes > 2 degrees) the anaesthetic bath should be renewed.

Following Anaesthesia:

After the procedure, fish should be recovered in a separate container prior to being gently returned to their rearing unit. Fish are monitored for recovery.

Anaesthetic baths must be disposed of in accordance with manufacturer recommendations and waste management regulations.

Fish populations should be monitored closely after all handling events. Mortality and morbidity should be assessed twice daily and all mortalities classified.

Some anaesthetic agents are subject to a withdrawal time. This is indicated on the prescription for the product. Fish ***must not be*** released or slaughtered for human consumption until after the withdrawal period has expired.

Details of the Operating Procedure:

4.30.1 ***TMS Anaesthesia:***

Equipment List

- Plastic tubs of appropriate size
- Anaesthetic stock solution or pre-weighed amount of drug to be used
- Sodium bicarbonate (baking soda) pre-weighed
- Supplemental air; tubing and airstones
- Personal protective equipment

The dose for TMS anaesthesia is 40 – 50 ppm.

Prepare sedative bath according to manufacturer's directions. Calculate the volume of water to be treated, and the appropriate amounts of drug and buffering agent (sodium bicarbonate for freshwater baths).

Dissolve the calculated amount of TMS needed in the holding tank. Mix well.

In freshwater, buffered TMS should be used for the safety of the fish.

- Check the pH of the water following TMS addition to determine the amount of sodium bicarbonate required. Add sodium bicarbonate till the solution has reached a pH of 7.0. Extremely hard water may not require buffering while soft water may require more than equal weight.
- Where measuring pH is not feasible, the standard rule of thumb is to add an equal amount of sodium bicarbonate to that of TMS added.

Aeration is supplied to both anaesthetic and recovery baths. Measure starting water quality parameters (dissolved oxygen, pH and temperature) and note the time of the addition of fish to the tank.

Test a small group of fish initially to detect any adverse response which could indicate either a species or strain sensitivity or a drug dosing miscalculation.

The maximum safe exposure time to TMS is 30 minutes.

Withdrawal time: Treated animals must not be slaughtered for human consumption or released to the environment for at least 5 days following the last treatment with this anaesthetic at water temperatures of 10° C or greater or at least 21 days at water temperatures less than 10° C.

4.30.1.1 Juvenile Anaesthesia

1. Assemble all equipment required for procedure
2. Prepare TMS stock solution in a metering beaker
3. Measure the TMS stock solution into the 6L anaesthetic basin
4. Add 1 tablespoon of baking soda into the 6L basin
5. Add well water to reach the measuring line in the basin
6. Fill another bucket with water. This will be used as a recovery bucket. If the fish are being returned to the ponds via hoses, the recovery bucket is not necessary
7. Place an airstone into the anaesthetic bath, and another one into the recovery bucket and ensure that they are both working
8. Dip net a few fish into the anaesthetic bath
9. Check the fish's response to the anaesthetic. If it appears the anaesthetic is to strong then add more water
10. When the fish loses equilibrium, proceed with the procedure planned
11. Change the anaesthetic bath every 15 minutes
12. If using a recovery bucket, transfer the fish from the bucket back to the pond when the procedure is finished

4.30.1.2 MSDS Information:

TMS is generally considered to be of low hazard, nonetheless it should be handled with caution. It is important that TMS, and all pharmaceuticals be handled in a safe manner using normal precautions.

Avoid contact with skin, eyes and clothing. Use in well ventilated areas. Wear gloves, dust mask, safety goggles and protective clothing. Wash exposed surfaces well with soap and water after use. Wash contaminated clothing before re-use. Do not breathe dust.

May be harmful by inhalation, ingestion or absorption.

In case of contact with:
Eyes: immediately flush with plenty of water for at least 15 minutes.
Skin: immediately wash with soap and water
Inhalation: immediately remove to fresh air, if not breathing give artificial respiration or oxygen.
Ingestion: give copious quantities of water.

Medical attention should be sought, especially if any irritation persists.

4.30.2 ***Carbon Dioxide:***

This is not a veterinary approved anaesthetic drug.

The dose for carbon dioxide gas in water is 150-200 mg/l to produce a loss of equilibrium in salmonids. To safely induce anaesthesia, dose testing with close monitoring of fish response, may be required.

Carbon dioxide should be used with caution as it has a low margin of safety due to its acidifying effects on the water, which are stressful for the fish.

Deeper anaesthetic planes are unreliable with this chemical and hyperactivity is a common response.

Note: The acidification of bicarbonate or carbonic acid evolves CO_2, which is similar in producing a hypercapnic condition in the water similar to CO_2 gas. One difference is the unknown contribution of the added acid to the pH change of the water. A common field method of using CO_2 is to use Alka-Seltzer® tablets. This requires some degree of testing to determine the appropriate dose for the fish in use and will depend on several factors but the literature indicates that a dose of four tablets per 500 mL provides euthanasia[2]. Narcosis to enable humane euthanasia has been reported to be induced by using 2-3 tablets per litre[3]. However, the problems with this chemical method of anaesthesia (hyperactivity, low plane of anaesthesia, low margin of safety, etc) persist despite source of carbon dioxide.

[2] Agriculture Fisheries and Forestry — Australia (2002). Destruction Manual (Version 1.0). Australian Aquatic Animal Diseases Emergency Plan (AQUAVETPLAN), Edition 1, Agriculture Fisheries and Forestry — Australia, Canberra, ACT.
[3] Handlinger, J. Collection and Submission of Samples for Investigation of Diseases of Fin Fish. of Fin Fish. Australian and New Zealand Standard Diagnostic Procedures. 2001. Available for free download. from http://www.affa.gov.au/aquaplan

See the Broodstock Handling *SOP for full details of the anaesthetic system*

Set up the anaesthetic (clove oil) and euthanasia (CO_2) baths

1. Turn off the drain valves to the anaesthetic tank, the 2 kill tanks, the sorting table & all drain valve lines
2. Add 135ml of Eugenol solution to the anaesthetic tank
3. Fill the anaesthetic tank using the wash down hose from the brail pool standpipe
4. Fill the kill tanks from the standpipe at the hydraulic pump
5. Charge the kill tanks with CO_2 at 40 L/min (full flow) for 15 min then reduce
6. Turn on the hydraulic pump and the pescalator (check to ensure that the anaesthetic flow through the pescalator is 3 L /min (each revolution of the screw takes 20 s)
7. Turn on the anaesthetic pump to deliver to the pescalator
8. Crowd adults to pescalator intake
    a. Adjust speed and anaesthetic flow to ensure adults are "knocked out" sufficiently to handle
    b. At the end of the sorting period (noon) drain the anaesthetic tank and refill.
9. Fill the male and female kill tanks with water and add carbon-dioxide
10. When water has reached saturation reduce the flow
11. Fill the round tank with water and add eugenol mixed with ethanol
12. Add sodium bi-carbonate to the tanks as a buffer
13. Crowd the appropriate # of fish into the crowding channel below the pond (don't overfill the channel)
14. Start the fish screw and adjust the rotation to 2.5 rotations per/min (speed will be adjusted relative to the size of the fish and relative to the use of anaesthetic (slower if using anaesthetic). When the fish enter the pescalator, they are dewatered via screens
    a. Anaesthetic (Clove oil) is recirculated through the pescalator and is refreshed approximately every two hours
    b. The pescalator takes approximately 45-60 seconds to lift a fish from the channel to the top and onto the sort table. Therefore, the anaesthetic is sufficient to sedate them enough to handle for ripeness checks
15. Sort the ripe males and females into their respective kill tanks
16. If the adults are too active on the sorting table slow down the rotation of the screw
17. If they are too submissive speed up the rotation to allow easy checking as well as a speedy recovery of the adults sent back to the holding pond
18. When the adults in the kill tanks have succumbed, rotate the finger paddles and allow the adults to go onto the spawning tables

4.30.2.1 MSDS Information:

CO_2 is a colourless, odourless gas. CO_2 may cause asphyxiation if used in a closed space without adequate ventilation. Inhalation of carbon dioxide acts as a weak narcotic at high concentrations. Inhalation of high concentrations of carbon dioxide can cause headaches, reduced hearing acuity, changes in respiration and increased blood pressure and pulse and asphyxiation. Contents of cylinders are under pressure. Liquid and cold vapour may cause tissue freezing.

4.30.3 ***Clove Oil (Eugenol)***

Note: This is not a veterinary approved anaesthetic drug.

Read the MSDS prior to handling

Eugenol is not water soluble and must be mixed with ethanol prior to dissolving in water to be used as an anaesthetic. Prepare the eugenol stock solution by combining 1/3 eugenol oil with 2/3 ethanol

The anaesthetic tank has a volume of 2200 L.

Eugenol is used at a concentration of 60mg/L

Therefore add 35 mL stock solution to the anaesthetic tank for use int eh pescalator.

Refer to the procedures above (and the Broodstock Handling *SOP)* for use of the anaesthetic in the sorting process.

4.30.3.1 MSDS Information:

Not believed to present a risk at present.

Forms & Records:

References:

Canadian Council on Animal Care: Guidelines for use of fish for teaching and research (http://www.ccac.ca/en/CCAC_Programs/Guidelines_Policies/GDLINES/Fish/Fish%20Guidelines%20English.pdf)
CCAC Guidelines for Anaesthetics (http://www.ccac.ca/en/CCAC_Programs/Guidelines_Policies/GDLINES/Fish/Fish%20Anesthetics%20-%20ENG.pdf)
http://www.syndel.com/anesthetics/anesthetics.html
http://www.syndel.com/anesthetics/aquacalm_info_sheet.html
http://www.syndel.com/anesthetics/tms_info_sheet.html
Fish Handling Procedures

4.31 Euthanasia

Rationale: In the uncommon event where fish require euthanasia (i.e. pathogen/disease sampling, disease control strategy) the procedure shall be done in a humane manner and rapidly and irreversibly result in loss of consciousness. This SOP addresses section *2.1.3.5* of the General Principles of Fish Health Management.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

All methods of euthanasia must result in a rapid and irreversible loss of consciousness. *It should be noted that while it is recognized that many facilities engage in suffocation by bucket, this is currently considered to be an inhumane procedure by the CCAC.*

Fish handling should occur in a humane and stress free a manner. (See *Fish Handling Procedures*). All fish culturists and fish health staff are required to know how to euthanize fish in a humane manner.

Acceptable methods of euthanizing fish include:

- Overdose with an anaesthetic agent (see *Anaesthesia*).
- A sharp blow to the head for larger fish held out of the water.
- Cervical dislocation and destruction of brain tissue.

When fish are killed for disease control purposes, biosecurity procedures should be followed to minimize the risk of disease spread within and from the premises.

Anaesthetic baths are disposed of in accordance with manufacturer recommendations and waste management regulations.

Details of the Operating Procedure:

Adults are euthanized by a sharp blow to the head using a mini-bat

Juveniles are euthanized by suffocation or may be vacuumed out to the fishway and flushed to the fishway

Forms & Records:
Mort sheets

References:
Anaesthesia
Fish Handling Procedures

4.32 Chemicals & Disinfectants: Supplies and Storage

Rationale: Chemicals and disinfectants must be handled, stored and administered/used properly to be efficacious. Selection of a disinfectant will depend on several factors, including the spectrum of activity of the disinfectant, the nature of the surface being treated, and the cost, safety, and ease of use of the available disinfectants. This SOP addresses section *2.3.5* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that all chemicals and disinfectants are used and stored in a manner that is efficacious and safe. All staff handling chemicals must be aware of WHMIS requirements.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

All staff handling chemicals must be aware of WHMIS requirements.

The following personal protective equipment is to be used when required:

- Latex gloves or heavy rubber gloves (latex gloves must not be reused beyond a single day)
- Rubber boots
- Safety glasses
- Respirator
- Coveralls, lab coats or a change of clothes as required for biosecurity purposes.

Chemicals should be stored in a manner that ensures both worker safety and prolonged efficacy and expiration information should be consulted prior to use where applicable.

Chemicals should be stored as recommended by the manufacturer. Provisions should be made to control temperature extremes, UV exposure, ensure adequate ventilation, etc. Disposal of chemicals will occur on the expiration date or sooner if there are any indications of decreased efficacy or problems encountered during storage.

All unused or spent chemicals should be disposed of according to manufacturer's directions in compliance with local waste management regulations.

Disinfectants must be used as recommended by the manufacturer and stored in a manner to ensure worker safety and prolonged efficacy.

All equipment and tanks should be cleaned prior to disinfection to ensure efficacy and to reduce the amount of chemical needed.

Disinfectant concentrations should be maintained either by checking concentration (e.g. test strips or visual inspection) or regularly scheduled renewal of the product.

Details of the Operating Procedure:

4.32.1 ***Compressed Gas Cylinders Storage***

Compressed gas cylinders must be stored in a clearly identified, dry, well-ventilated storage area away from doorways, aisles, elevators, and stairs. “No smoking” signs are clearly posted in the area. Store cylinders in the upright position and secure with an insulted chain or non-conductive belt and secure the protective caps.

Compressed gas cylinders (full and empty) that are not currently in use are stored behind building #1 in the bottle rack storage area. O_2 containers in use are stored in the lab and are securely chained to the wall. Those cylinders stored outside the building are chained and clear signage is installed. These are always stored with caps and are labelled as empty or full.

Gas cylinders used for adults are stored in the storage area.

The area is well ventilated and cylinders are on a fireproof surface. The enclosure is tamper-proof and cylinders are protected from contact with ground, ice, snow, water, salt, corrosion, and high temperatures.

Oxygen and fuel gases are stored separately.

Indoors, oxygen must be stored at a distance from fuel gas cylinders of at least 6 metres (20 feet), by a wall at least 1.5 m (5 feet) high, or in cylinders rated for 1.5 hour fire resistance

There is always an extra full cylinder of oxygen and CO_2 on hand when it is being used regularly during adult handling and fish transport. Try to keep no empties on hand as they get charged rental.

Cylinders are moved by truck. The truck has a rack installed which securely holds cylinders from rolling or accidental damage.

The cylinder should not be “cracked” until after the regulator has been attached properly and cylinder is secured in place.

4.32.1.1 MSDS Information:

CO_2 is a colourless, odourless gas. CO_2 may cause asphyxiation if used in a closed space without adequate ventilation. Inhalation of carbon dioxide acts as a weak narcotic at high concentrations. Inhalation of high concentrations of carbon dioxide can cause headaches, reduced hearing acuity, changes in respiration and increased blood pressure and pulse and asphyxiation. Contents of cylinders are under pressure. Liquid and cold vapour may cause tissue freezing.

4.32.2 ***Vidalife/Poly-Aqua***

Vidalife is the trade name for a specially formulated water conditioner for use in hatchery and transport tanks, on handling equipment and handling surfaces. Vidalife can be used whenever fish are handled or moved. Used as described, Vidalife will help protect fish by preserving the natural mucus layer as it purportedly forms a coating on contact surfaces to reduce friction and abrasion

when handling. Vidalife helps make a protective barrier between fish and handling equipment and is suggested to reduce the toxicity of heavy metals. Vidalife may help reduce stress and abrasions during any handling process and therefore, it may reduce vulnerability to pathogens that may affect a fish.

Vidalife is stored in the lab in a 20 L pail

4.32.2.1 MSDS information:

Vidalife contains polyvinylpyrrolidone and tetrasodium EDTA, both of which are suspected to be carcinogenic. Irritation may result from eye contact or ingestion.

4.32.3 ***Chloramine-T***

Chloramine-T is a disinfectant used for surface bacterial infections including bacterial gill rot.

Chloramine-T is stored in the lab in a 20 L pail

When handling Chloramine-T a respirator and rubber gloves are worn.

4.32.3.1 Chloramine-T MSDS information

Chloramine-T powder can cause burns or sensitization on skin contact and sensitivities upon inhalation; it is injurious to eyes and is harmful if swallowed. Staff should review WHMIS information prior to handling this product and employ appropriate personal protective equipment.

4.32.4 ***Parasite-S***

Parasite-S™ is the trade name for an anti parasitic, formalin-based solution that is commonly used against parasitic, fungal and bacterial gill infections. It is also a standard disinfectant used in hatcheries for the prevention and treatment of egg fungal infections (see *Egg Fungal Treatments*).

The normal dilution of formalin for treating fish is 1:6000 or 167 ppm. This concentration is achieved by combining 17 mls of Parasite-S per 100 litres of water. Exposure is normally 30 – 60 minutes daily, and may be done on consecutive or alternating days for three treatments in total.

Parasite-S is stored in the incubation room in a dedicated storage cabinet. Safety information is clearly marked and the cabinet is clearly labelled.

Cautions:

Formalin should never be added to water containing fish without first diluting it and then mixing it in thoroughly to avoid 'hot spots'.

Formalin should not be used if :

- Dissolved oxygen of the water is <5ppm
- The water temperature is >27^{o}C

- Heavy phytoplankton growth is present.

4.32.4.1 Parasite-S MSDS information:

Formalin must be handled with care. It is harmful if inhaled, and can seriously irritate eyes and skin after contact. Formalin should only be handled in well ventilated areas, preferably while wearing a respirator and safety glasses. Staff should review WHMIS information prior to handling this product and employ appropriate personal protective equipment.

Parasite-S contains methanol which inhibits the formation of paraformaldehyde, a white precipitate that is extremely toxic to fish. Even so, formalin should be stored in the dark above 10^{o}C and not allowed to freeze. If Parasite-S is allowed to freeze, it should be discarded due to the rapid formation of paraformaldehyde.

The toxicity of formalin increases as temperature increases. In very soft water, concentrations of formalin should not exceed 25 ppm. Concentrations greater than 250 ppm may cause severe gill damage and should not be used on salmonids.

Formalin is a gill irritant and thereby reduces gas exchange. This is especially of concern as formalin is commonly used when fish gill function may already be compromised. Additionally, formalin is a reducing agent that absorbs oxygen from the water. Therefore, the safest course is to treat with formalin at the time of day when the water temperature is at its lowest, and to provide supplemental oxygenation via airstones. The fish should be closely monitored during treatment for signs of respiratory distress (increased opercular movements or gasping at the surface) and the treatment terminated if needed.

4.32.5 ***Hydrogen peroxide***

Hydrogen peroxide (H2O2) is a clear, colourless liquid. It is a weak acid with strong oxidizing properties and is therefore a powerful bleaching agent that has found use as a disinfectant and as an oxidizer.

Hyperox is used as a source of hydrogen peroxide. Hyperox is stored in the in a 20L pail

4.32.5.1 MSDS information:

Hydrogen peroxide should be stored in a container made from a material that doesn't react or catalyse the chemical. Numerous materials and processes are available, some stainless steels, many plastics, glasses and some aluminum alloys are compatible. As peroxide is a strong oxidizer it should be stored away from fuel sources. Apart from obvious fire risks, peroxide vapour can react with hydrocarbons such as alcohols to form contact explosives.

Because oxygen is formed during the natural decomposition of the peroxide, the resulting increase in pressure can cause a container (e.g. made of glass) to shatter. Peroxide should be kept cool, as peroxide vapour can detonate above 70C.

At higher concentrations, hydrogen peroxide, if spilled on clothing (or other flammable materials), will preferentially evaporate water until the concentration reaches sufficient strength, then

clothing will spontaneously ignite. Leather generally contains metal ions from the tanning process and will often catch fire almost immediately.

Concentrated hydrogen peroxide (>50%) is corrosive, and even domestic-strength solutions can cause irritation to the eyes, nose, throat, mucous membranes and skin. Contact may cause irreversible tissue damage to the eyes including blindness. Swallowing hydrogen peroxide solutions is particularly dangerous, as decomposition in the stomach releases large quantities of gas (10 times the volume of a 3% solution) leading to internal bleeding. Severe pulmonary irritation may result from inhalation of fumes from solutions of over 10%.

Immediately flush eyes with water for at least 15 minutes, lifting the upper and lower eyelids intermittently. See a medical doctor or ophthalmologist immediately. On skin contact, wash with plenty of soap and water. Get medical attention if irritation occurs and persists. If ingested, rinse mouth with water. Dilute by giving 1 or 2 glasses of water. Do not induce vomiting. See a medical doctor immediately.

4.32.6 ***Sodium chloride***

Sodium chloride is used for transporting fish to marine pens.

Sodium chloride is stored in dry storage in the mezzanine in the shop

4.32.7 ***TMS***

TMS is a chemical used to provide anaesthesia to fish, thereby reducing the degree of stress associated with many fish culture procedures.

TMS is stored in the fridge in the lab

4.32.7.1 MSDS information:

TMS is generally considered to be of low hazard, nonetheless it should be handled with caution. It is important that TMS, and all pharmaceuticals be handled in a safe manner using normal precautions.

Avoid contact with skin, eyes and clothing. Use in well ventilated areas. Wear gloves, dust mask, safety goggles and protective clothing. Wash exposed surfaces well with soap and water after use. Wash contaminated clothing before re-use. Do not breathe dust.

May be harmful by inhalation, ingestion or absorption.

In case of contact with:
Eyes: immediately flush with plenty of water for at least 15 minutes.
Skin: immediately wash with soap and water
Inhalation: immediately remove to fresh air, if not breathing give artificial respiration or oxygen.
Ingestion: give copious quantities of water.

Medical attention should be sought, especially if any irritation persists.

4.32.8 ***Ovadine™***

Ovadine is a specially buffered, non-corrosive, aqueous iodine solution used by fish culturists as a general disinfectant on equipment, tanks, nets, hands and clothing in hatcheries and at farm sites. It is also used to disinfect eggs. It is a fast acting disinfectant that has been shown to be effective against many gram-positive and gram-negative bacteria and fungi.

Ovadine is stored in the lab in 20 L pail

Weak solutions of Ovadine are diluted with several volumes of water before discarding. Strong solutions are neutralized with sodium thiosulphate prior to discarding. Large volumes of Ovadine will be neutralized (with sodium thiosulphate) and disposed of in a landfill.

4.32.8.1 Ovadine MSDS Information

Synonym- 10% Povidone iodine solution

There is no evidence of any hazard associated with inhalation of Ovadine solution. There is no evidence of any adverse effects of ingestion or skin contact with Ovadine. Ovadine solution is classified as practically non-toxic. Even so, eye and skin protection is advised .

Storage in high temperatures results in a loss of available iodine in solution.

4.32.9 ***Virkon***

Virkon is the trade name of a disinfectant chemical containing peroxygen compounds, a surfactant, organic acids and an inorganic buffer system.

Virkon is stored is the lab

4.32.9.1 Virkon MSDS Information

Rubber gloves, a dust mask (stored in the cupboard above the counter) and protective eyewear are worn when measuring out Virkon powder as per manufacturers requirements. Good ventilation should be used. Care should be used when measuring out Virkon powder to avoid the generation of airborne dust which can be irritating.

Virkon may be very irritating to the skin, eyes and mucus membranes and should be handled with a degree of caution. If contact is made with skin, rinse well. If eye contact is made, rinse eyes well with water for at least 165 minutes and obtain medical attention. Ingestion may result in severe irritation tot eh throat, digestive tract and stomach. Do not induce vomiting, drink plenty of water (or milk) and seek immediate medical attention.

4.32.10 ***Isopropyl alcohol***

Isopropyl alcohol is used for sterilizing sampling instruments in the laboratory.

4.32.10.1 Isopropyl alcohol MSDS Information

Synonyms: 2-propanol, isopropanol, dimethylcarbinol

Isopropyl alcohol is a colourless liquid with the odour of rubbing alcohol. It has a low flash point and is extremely flammable and flames are virtually invisible. Water is generally not effective in extinguishing fires, carbon dioxide or dry chemical extinguishers are appropriate fire fighting media.

Storage is incompatible with strong oxidizing agents (nitrates, perchlorates, peroxides) which may increase the risk of fire and explosion.

At high concentrations, isopropyl alcohol may cause mild irritation of the upper respiratory tract, drowsiness, ataxia (inco-ordination) and deep narcosis. Direct eye contact can cause severe irritation, flush with water for 15 minutes and seek medical attention. Brief skin exposure is not generally irritating and absorption through the skin does not readily occur so toxic doses are unlikely. If skin contact occurs, wash with soap and water and remove contaminated clothing. Ingestion may lead to drowsiness, gastrointestinal pain, cramps, nausea, vomiting and diarrhea. Seek medical attention. Unconsciousness and death may occur following large doses. Vomiting, by having individual drink large quantities of water, should be induced if ingested. Lethal dose for humans is approximately 130 grams.

Store in tightly closed, electrically grounded containers in a cool area and away from sources of heat, sparks or flame. The storage area should have adequate ventilation and have no source of heat or sparks. Fans and other electrical motors should be spark resistant.

Use minimal quantities in designated areas with adequate ventilation and away from sources of heat or sparks. User fire resistant containers and store closed in a grounded, fire resistant cabinet. Alcohols are incompatible with acids and strong oxidizing agents. Store separately from these.

If small amounts are to be disposed down a sink, flush with ample water to prevent the accumulation of flammable vapours.

Chemical splash goggles should be worn to protect eyes from liquid or concentrated vapours. Gloves should be worn to prevent exposure of skin.

4.32.11 ***Ethanol***

70 – 95% ethanol (also known as ethyl alcohol or EtOH) may be used as surface disinfectant for instruments (i.e. spawning knives, egg picking tweezers, dissection equipment, etc.) or lab benches. Note: 70% is commonly used due to the rate of evaporation at higher concentrations.

Ethanol can be stored in sealable glass or plastic containers when not in use, and poured into a small beaker for instrument tip disinfection when required.

For lab bench surfaces, 70% ethanol may be transferred into a plastic spray bottle for use. It should be sprayed to coat the desired area of a clean bench top, left for roughly one minute contact time, then the excess may be wiped off with a paper towel.

Ethanol is stored in the lab on the shelf in a 20 L pail

4.32.11.1 Ethanol MSDS Information

Although Ethanol is relatively stable, it is hygroscopic and substances to be avoided include strong oxidizing agents, peroxides, acids, acid chlorides, acid anhydrides, alkali metals, ammonia, moisture. It may forms explosive mixtures with air.

Ethanol may cause skin and eye irritation. Ingestion can cause nausea, vomitting and inebriation; chronic use can cause serious liver damage. Note that "absolute" alcohol, which is close to 100% ethanol, may nevertheless contain traces of 2-propanol, together with methanol or benzene. The latter two are very toxic, while "denatured" alcohol has substances added to it which make it unpleasant and possibly hazardous to consume.

Extreme caution should be exercised if working with ethanol near flame sources as it is highly flammable and the flame may be invisible in well lit areas.

Forms:
Chemical inventory records

References:
WHMIS Material Safety Data Sheets
Hyperox™ data sheet: http://www.antecint.co.uk/Main/hypox.htm
Virkon™ data sheet: http://www.antecint.co.uk/MAIN/vkuse.htm
Ovadine™ data sheet: http://www.syndel.com/d_p_f_s/Ovadine_info_sheet.html

4.33 Equipment disinfection

Rationale: Equipment is to be kept clean at all times to limit pathogen spread. Movement of equipment between sites provides easy access for pathogens to migrate to facilities in different geographical regions. This should be avoided to reduce the risks associated with pathogen transfer. This SOP addresses section *2.2.7* of the General Principles of Fish Health Management.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

4.33.1 ***Between sites:***

Each site should have designated equipment that should not be shared with other sites.

On the rare occasion when equipment must be shared with other sites it should be cleaned, disinfected and dried prior to leaving the site and prior to returning to the originating site.

Each site should have designated raingear that is not to be transferred between sites.

Vehicles and vessels used between multiple sites should be cleaned and disinfected prior to use at another site. Acceptable methods include 'fallowing', drying, and ultraviolet light and chemical disinfectants.

4.33.2 ***Within the site:***

Each rearing unit should have designated equipment that should not be used in other rearing units.

After use, equipment such as dip nets, buckets and feeding equipment should be cleaned, disinfected, dried and put away in the proper location.

When equipment must be shared between rearing units (for example: large objects such as grading tables), it should be cleaned, disinfected and dried between uses on different fish groups.

The wastes from cleaning operations should be managed in a manner that minimizes pathogen spread and environmental damage. The use of high pressure water generates aerosols that can spread pathogens easily. Caution should be exercised whenever high pressure water is being used for cleaning of any type.

In the freshwater environment, holding units should be cleaned, disinfected and dried between groups of fish housed in the facility. Tanks should be cleaned whenever organic matter has accumulated or algal growth becomes problematic. High pressure water is often insufficient to remove biological matter and surfaces may require manual scrubbing.

At all sites holding units should be cleaned and disinfected prior to housing different groups of fish. Acceptable methods of disinfection include fallowing, drying, ultraviolet light and chemical disinfection.

4.33.3 ***General Disinfectant Protocols:***

Disinfectants are chosen based on the anticipated degree of microbial killing required, the nature of the surfaces involved (i.e. rubber versus stainless steel versus concrete), and the cost, safety and ease of use of the chemical. Selection of appropriate disinfectants should be made by the site management in consultation with the Veterinarian.

Products should be used according to manufacturer's directions.

Disinfectant concentrations should be maintained either by checking concentration (e.g. Virkon™ using test strips) or regular renewal of the product (e.g. Ovadine™ dips replaced twice weekly or when indicated by a colour change).

Disinfectants should be disposed of according to manufacturer directions in such a manner that meets the requirements of waste management regulations.

Details of Operating Procedures:

4.33.4 ***Equipment Disinfection Protocol:***

Equipment cleaned and scrubbed with water prior to disinfection to remove all visible organic matter.

Clean equipment is disinfected with Hyperox according to the manufacturer's direction on the container.

Ten minutes of contact time is required for successful disinfection with most products. Some products may require a greater degree of contact time. Follow the manufacturer's recommended guidelines. Equipment will not be left in the disinfectant bath indefinitely as this can result in deterioration of equipment.

All disinfected items should be rinsed with fresh, clean water before being put away in their proper storage location. Inadequate attention to rinsing can leave residual disinfectant behind that can be harmful to fish.

Equipment should be allowed to dry before re-use.

4.33.5 ***Tank Disinfection Protocol:***

Tanks and ponds are rinsed of debris using a high pressure fire hose. Tanks are scrubbed if needed and smaller containers may be disinfected with Hyperox. Containers are rinsed with fresh, clean water, and allowed to dry after the removal of fish and prior to the introduction of new fish.

Heath trays are scrubbed by hand and rinsed with house pressure water before being disinfected. After disinfection, trays are rinsed with clean water and left to dry.

Adequate contact time for the disinfectant is allowed (please review this for the disinfectant concentration being used – a minimum contact time of 10 minutes is standard).

Disinfectant is also sprayed onto any tank-associated equipment (inlet pipes, tank rims, stand pipes, etc)

Where possible, a fallow period of at least one week between fish groups should be allowed.

4.33.6 ***Foot Bath and Foot Mat Disinfection Protocols:***

Footbaths filled with Virkon are used in the fall during incubation. Virkon concentration is checked using a testing kit and is changed or topped up as necessary.

Dispose of excess solution into a drain that goes to municipal sewage or dispose to ground.

Record foot mat/bath change on the footbath log.

Monitor footbath concentration at a minimum of twice weekly and change when the concentration has declined below effective levels or when it appears heavily soiled.

4.33.7 ***Instrument Disinfection Protocol:***

Isopropanol is used as surface disinfectant for dissection instruments.

Isopropanol can be stored in sealable glass or plastic containers when not in use, and poured into a small beaker for instrument tip disinfection when required. The beaker needs to be wide enough or heavy enough to resist tipping over when handled instruments are placed tip-down inside it.

For lab bench surfaces, 70% ethanol or isopropanol may placed be into a plastic spray bottle for use. It should be sprayed to coat the desired area of a clean bench top, left for roughly one minute contact time, then the excess may be wiped off with a paper towel.

Organic matter should be brushed off with a small scrub brush prior to instrument immersion in ethanol to ensure effective disinfection.

Instruments should be immersed for at least one minute prior to re-use to allow sufficient time for disinfection.

Pass the instrument tip through a flame to burn off the alcohol and allow the tip to cool for 5-10 seconds before use.

Cautions:

- during flaming, the instrument tip showed be angled downward to prevent burning ethanol from running down the instrument and coming in contact with operator
- burning alcohol may not be visible beware replacing a flaming tip back into the alcohol.

4.33.8 ***Ovadine™***

For use as a **general disinfectant** for dips for equipment or footbaths, a 250 ppm available iodine solution is made by diluting 25mLs of Ovadine™ to 1 litre of clean water.

A change in the solution colour from dark brown to light yellow indicates a loss of activity.

For use as an **egg disinfectant**, a 100 ppm available iodine solution is made by diluting 10 mLs of Ovadine™ to 1 litre of clean water. (see Egg Disinfection)

Ovadine™ may be stored at room temperature (20 – 30^{o}C) for periods greater than two years if containers are kept tightly sealed and away from direct sunlight.

4.33.9 ***Virkon™***

To create a 1:100 solution (1%) of Virkon™ add 100 mg of disinfectant to 10 litres of fresh water. The pink colour is an indicator of efficacy; concentration test strips are also available.

Footbaths are replenished every 2 – 3 days depending on traffic or when heavily soiled or when the colour changes.

Nets, or other surfaces which are difficult to remove organic matter from, are soaked in Virkon™ solution for 20 – 30 minutes followed by rinsing with water.

Virkon™ should be stored at room temperature. For the powder, a 2.3% loss in activity will be seen following 36 months in storage. For a 1% working solution, a 10% loss of activity will be seen after 7 days in 350 ppm hard water.

4.33.10 ***Hyperox™***

To create a 1:128 solution (0.8%) of Hyperox™ add 80 mLs of disinfectant solution to 10 litres of fresh water.

Using test strips to assess the concentration of Hyperox™ disinfectant solutions:

Dip a Hyperox™ test strip into the disinfectant solution for two seconds; remove and allow excess solution to run off. Immediately read the dilution rate off the colour chart on the test strip container. The target value is 0.8%. Change the disinfectant solution if the concentration is lower than this. Also change disinfectant solution if it appears dirty or contains excessive organic matter.

Hyperox contains 5% peracetic acid and 25% hydrogen peroxide with a surfactant. Hazardous decomposition products are oxygen and acetic acid. Hyperox is rapidly degraded in soil/water by hydrolysis, decomposition and reduction. Acetic acid formed by decomposition is biodegradable. The product will be diluted and discarded.

Forms & Records:

References:

Hyperox™ data sheet: http://www.antecint.co.uk/Main/hypox.htm
Virkon™ data sheet: http://www.antecint.co.uk/MAIN/vkuse.htm
Ovadine™ data sheet: http://www.syndel.com/d_p_f_s/Ovadine_info_sheet.html

4.34 Vaccine handling, storage and administration

Rationale: Vaccines must be handled, stored and administered properly to be efficacious. This SOP addresses sections *2.1.3.4* and *2.3* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that vaccination is carried out in a manner that is efficacious.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Fish showing signs of illness should not be vaccinated and fish should not be vaccinated at water temperatures of less than 1°C. Vaccination should also be avoided during smoltification and the four weeks prior to seawater entry

4.34.1 ***Vaccine Storage:***

The vaccine should be stored according to manufacturer's directions. Generally, it is kept refrigerated (2-8°C) in transport and storage and is protected from light and freezing.

Once opened, a bottle of vaccine should be used within 24 hrs. Any open vaccine older than 24 hrs must be discarded.

The vaccine should be a uniform cloudy suspension. If the vaccine appears abnormal it should not be used.

The lot number of the vaccine should be noted in the records and the expiry date checked prior to use; expired vaccine is discarded.

4.34.2 ***Before Vaccination:***

Fish should be taken off feed for a maximum of 72 hours prior to vaccination. Determination of the time off food includes consideration of fish size, diet, water temperature and existing knowledge of gut emptying times.

A risk assessment should be conducted by fish health management prior to the procedure. This will include analysis of the health status of the stock, their age and size, previous disease history, current morbidity, mortality rate and important production parameters such as feed conversion ratio. If there are any indications that the health of the fish may be compromised, the veterinarian should be consulted prior to proceeding.

Vaccination should not occur if water quality is questionable.

Once it has been determined that fish are healthy enough for vaccination, large enough to respond to the vaccine and the environmental parameters are acceptable, the process can proceed.

A checklist of required equipment should be available and checked off prior to starting fish handling.

4.34.3 ***During Vaccination:***

Vaccines are to be used according to manufacturer's directions.

Fish should be handled gently to minimize stress (see common *Fish Handling Procedures*).

Water quality in the bath should be monitored closely for the duration of the procedure, particularly dissolved oxygen and temperature, supplemental oxygen is bubbled into the vaccine bath to maintain optimum dissolved oxygen levels.

Fish should be captured in a dip net or bucket, holding water is allowed to drain from the container (fish may be weighed at this point). Numbers of fish in the net or bucket should not be excessive; fish should be closely monitored for signs of injury during the procedure.

The fish should be immersed in the vaccine solution for 30 seconds. The fish are then removed from the dilution, drained and returned to the holding unit and monitored closely for signs of injury from handling.

This procedure is repeated until 100 kg of fish per litre of diluted vaccine have passed through the bath.

The vaccine bath must be discarded according to manufacturer recommendations and local waste management regulations. Any unused, open vaccine is discarded after 24 hours.

All equipment used should be cleaned, disinfected and put away in its proper place (see *Equipment disinfection*)

Fish should be fed the day following vaccination if their behaviour and appearance is normal. Fish should be monitored closely for two weeks following the vaccination procedure for signs of illness. Fresh mortalities should be closely examined and sampled.

Details of the Procedure:

Before vaccination is to take place starve fish for 3-4 days.

After vaccination the holding time is:

- 21 days prior to release
- 14 days @ 10 degrees prior to transfer to saltwater

4.34.4 ***Set-up & Operation of the Vibrio Bath***

Materials Required

- □ Vibrio Vaccine (1 liter per 100 Kg of fry to be vaccinated)
- □ 50 liter holding containers
- □ dewatering baskets
- □ Oxygen bottle complete with pressure gauge, flow meter, & air lines
- □ air stones
- □ stopwatch
- □ dip net
- □ Clean well water
- □ Ice packs
- □ Pond seine net
- □ 3 person crew
- □ Pencil & Paper (to keep track of Kg of fry to each tub)
- □ **NOTE: (**if moving fry from ponds 15-17 to the concrete ponds) have transport tank ready
- □ Transport line
- □ Scale

Read MSDS sheet on the Vibrio Vaccine and take the appropriate H&S measures to ensure the safe handling of this product. Wear eye protection & latex gloves while working with the vibrogen (mixing)

1. Set up equipment as close as possible to fry that are to be vaccinated
2. Set up the anaesthetic tub on the scale
3. Mix 1 liter of the vaccine with 9 liters of water into a holding container (normal mix will be 3L vaccine to 27L well water) for 1 container, mix thoroughly
4. Assemble the air system by connecting the airline to an airstone. Place the airstone in the vaccine bath along with an ice pack to keep the solution cool while in use
5. Place the dewatering basket in the holding container with the vaccine and mark the holding container at the height of the vaccine level
6. Now add a measured 5 & 10 kgs of fry and mark the level of these heights also
7. Bubble oxygen into the vaccine at a flow rate of 2L per minute

NOTE: (if moving fry from ponds 15-17 to the concrete ponds) have transport tank ready complete with oxygen & water set at desired tank load level

8. Ensure that the Transport line is set up to deposit vaccinated fry to receiving pond

   Once the vaccination tubs are ready, the procedure may proceed.

9. Using a seine net, crowd the fry to be vaccinated
10. Designate staff duties as below:
    a. Designate one person to be the timer/recorder to control the operation and ensure that the fish are in the vaccine for the 20-30 seconds that is required. This person

will also monitor the oxygen and temperature of the vaccine and will put the dewatering tub into the vaccine tub.

b. Designate a second person to operate the dipnet. This person will collect the fish that are to be vaccinated and pour the water drained fish into the dewatering tub that is placed in the vibrio tub.

c. Designate a third person to remove the dewatering tub with the fish in it from the vaccine when indicated to do so, this person will return the fish to the rearing container

11. Dip net fish from the pond
12. Briefly dewater the fish and then pour them into the perforated dewatering tub in the vaccine bath filling the container to the designated volumetric line
13. Record the weight of fish added to the vaccine bath
14. Time for 30 seconds
15. Lift the dewatering tub from the bath and allow to drain
16. Transfer the vaccinated fish into the receiving pond
17. The dewatering basket must be placed back into the holding container before more fry can be added
18. When doing large amounts of fry it is recommended that 2 containers be used to help speed up the process
19. Repeat the last 3 steps until done
20. Discard used bacterin after 100 kg of fish have been processed & start with fresh if needed.

Note: do not let the vaccine warm up

Note: remember that your crew members should be rotated to relieve the back and arm strain that is associated with the lifting of all the weight that you will handled during this process.

Note: (if moving fry from ponds 15-17 to the concrete ponds) have transport tank ready complete with oxygen & water set at desired tank load level (normally at 100 Kg load

Forms & Records

Vibrio Vaccination Form.doc

References:

Equipment disinfection
Fish Handling Procedures

General Practices and Procedures

4.35 Predator exclusion

Rationale: Predator interactions with fish can result in stress, injury, and death. These interactions create stressful situations which predispose fish to disease and decrease productivity. Predators and scavengers can introduce pathogens to the fish and predator damage to infrastructure can lead to fish escape. This SOP addresses section *2.2.4* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that predators do not gain access to fish holding units.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

4.35.1 ***Infrastructure***

The use of predator exclusion devices is critical. In the freshwater environment indoor facilities, fully fenced sites, covered fish holding units, the use of bird netting on fish holding units and screens on effluent drains are some predator exclusion options.

Details of the Operating Procedure:

A custom fit pool blanket is used to cover Sarita broodstock to reduce any distractions from staff walking around these ponds. In recent years, bears have become a nuisance and often get into these ponds - primarily the deep Nitinat Chinook concrete raceways.

In 2004, the entire adult holding area was enclosed with chain link predator fencing, effectively stopping the bear problems altogether.

Predator netting is placed on the ponds as necessary. Newly ponded fry are protected with predator netting or string with flagging tape to deter birds.

In the fall, prior to adults being brought into the facility, an electric fence is erected *around much of the site. It extends from building 1 (pond 4) through and around the tubs. This is reduced to a smaller area as the season progresses.*

4.35.2 ***Procedures***

Facilities should be checked daily for signs of predators. Any damage to holding units is to be repaired as soon as possible.

Feed should be stored and distributed in a manner that does not increase attraction to scavengers and predators. (See *Feed, Feed Storage, & Feeding Practices*) Spilled feed is cleaned up immediately.

Household refuse should be properly contained prior to removal from the site.

Mortalities should be examined regularly for signs of predator attack (See *Mortality Classification*)

Details of the Operating Procedure:

Garbage is placed into the walk in cooler. The cooler is shut down for 3 of the year (between June and August) and any household garbage is placed into the aeration tower in totes during this period

4.35.3 ***Contingency plan***

When exclusion methods have failed and all other options have been exhausted, a conservation officer should be called and permits must be obtained in order to trap and relocate or terminate predators. Predator destruction must be done in as humane a manner as possible. The predator population must not be put at undue risk.

Details of the Operating Procedure:

Staff possess trapping and firearms licences and will destroy nuisance predators if required.

4.35.3.1 Predator Control Guidelines (February, 2005)

Until a regional policy is implemented for DFO hatcheries, these guidelines will be followed at Nitinat River Hatchery to deal with predators that pose a direct risk to cultured fish that are within the hatchery owned and controlled areas. These guidelines will use best practices, given the information that is available and the limits that staff has to work under.

POLICY
It is the policy at Nitinat River Hatchery to do as much as possible to deter predators from entering fish rearing areas of the site. It must be recognised however, that the ability to do preventative measures is limited by the budget and capital projects priority list that is managed and allocated by Area and Regional Managers and engineering staff. Health and Safety concerns must also be considered as well as ability to work in the area where control measures have been put into place.

METHODS

The strategy for dealing with predators is based on the following principles:

1. Avoidance: If possible, do not engage in activities that will attract predators.

- This includes not leaving garbage, fish food, fish morts, or other attractors out.
- Keep gates closed and pond covers secure when left unattended.
- Wash down work areas thoroughly.

2. Deterrence: If possible, erect predator fencing, electric fences, and netting:

- Ensure maintenance is done on fences, nets, and screens
- Plan the work to rear fish in the most controlled and secure areas
- Keep pond levels low so that herons can't bend over to get fish, but not so low they can stand
- Try scare techniques such as sprinklers, noise, flags, dogs, etc., (although these methods have not been proven effective for more than a short time).
- Remove perching structures

3. Termination: If the other strategies fail or cannot be achieved, then *permitted* predator animals will be legally killed under permit by qualified staff or contractors.

I. Shooting of migratory birds "actively engaged in destroying fish stocks" (Environment Canada Permit).

- This allows for control of species under the Federal Migratory Bird Regulations such as Ducks & Mergansers. This permit does not allow the killing of certain Provincially Protected species or Species at Risk, in particular, Raptors or Great Blue Herons.

II. Shooting or lethal trapping of Weasel, Otter, and Mink (Provincial Permit) Black Bears are also included in the predator permit, but Conservation Officer Service should be notified as first responder

III. All Wildlife posing a risk to worker or public safety, (bears, cougars), should be reported to the local Conservation Officer Service.

4. Live Trapping: Live trapping and release has been suggested as an alternative to lethal methods.

Although this option avoids the unpleasant task of destroying an animal, it may also not be something that would be in the best interests of the animal or the ecosystem that it is released to. Dumping one more predator into another watershed on top of a food chain that already exists, could imperil the fish & wildlife that already occupy that niche. Further, the Provincial regulations require that an application be made to MWLAP before any transplants of wildlife be done (as per the Wildlife Act). Transplants are not granted easily, as concerns and consideration is seriously given to the ecosystem where the animal is going. For those species that a permit has been granted to lethally trap, the Ministry does not have a concern for the viability of that species or population, otherwise they would not have granted the permit.

It must be noted that transplanting without a permit is against the law.

If non-permitted species are live trapped (domestic cats, raccoons), then they should be released, as they do not pose a serious predator problem. Domestic feral cats can be taken to the local SPCA to be dealt with there, and other species released in the local area away from the hatchery grounds.

5. Predator Control Using Firearms

A. All employees must follow the current "Firearms Policy for Non-Enforcement Staff" December, 1998. It should be noted that a new policy is in draft and in final stages of review and approval and some changes are anticipated. Mandatory requirements in the policy are:
    - All employees that carry and/or use a firearm must possess a valid PAL (Possession and Acquisition Licence).
    - Must have taken and passed the Fisheries & Oceans Firearms for Non-Enforcement staff course.
    - Must have qualified in annual re-certification shoot.

B. If a predator must be killed using a firearm, it should be done in a safe and humane manner. Consideration must be given for other workers and members of the public that may be in the area. The proper firearm and ammunition must also be used depending on the species and conditions encountered.

    - When dispatching a predator, ensure that the general area is clear of members of the public.
    - Warn other employees that are working near that you have to shoot a predator, to stay clear of the area, and be prepared for the noise from the gun.
    - Approach the scene with care and be aware of potential hazards to footing, damage to structures or equipment from missed shot, and the potential for ricochets.
    - When animal/bird has been killed, remove it from the area with discretion and record and dispose of it as per the instructions on the predator permit.

6. Predator Control Using Traps: All personnel that are required to do lethal trapping must have a B.C. Trappers Licence

Alternative is to have a licensed trapper come onto the site and set the traps.

At Nitinat Hatchery, a licensed trapper has a permit to trap in the surrounding Nitinat watershed, and has done this for many years keeping control of Wolf, Mink, Otter, and Beaver populations. His name is Peter Olson and can be contacted at: 749-3948. In addition, Ditidaht Band has an elder (Mike Thompson) who has expressed interest in the furs and will also pursue trapping around the hatchery with guidance from Peter Olson and trained hatchery staff. The Ditidaht Band contact regarding trapping is Fred Seiber who can be contacted at 745-3333.

All lethal trapping will follow the instructions given on the trappers education course, (types of traps to use, methods, etc.). This information will be incorporated into these guidelines when available.

1. All animals lethally trapped will be recorded and disposed of as per the predator permit.
2. All set traps will be "flagged" in close proximity to the trap with a pole and flagging tape to warn workers, (and all workers will be educated about potential traps prior to setting).

3. Traps will not be set during day-light hours when workers must operate in the area, or at times when qualified staff are not on-hand to deal with the animal (weekends, holidays, etc,).
4. Live trapping and subsequent destroying of permitted predators by drowning or shooting is an approved method until more information and alternate methods can be found (depending on the information gained from the trappers course).

Any concerns raised in regards to predator control at Nitinat River Hatchery should be directed to the Hatchery Manager. Staff that are engaged in the actual work of controlling predators are doing so as part of their duties to enhance and protect valuable fish stocks under their care at the hatchery. They must not be hampered directly or indirectly in their task by other staff. Alternately, staff performing predator control must consider all alternatives and risks before using lethal means. They must also exercise a high degree of discretion and consider the thoughts and feelings of other staff members when having to deal with these situations.

Forms & Records


Predator Summary Sheet.xls


Bear-Encounter STEPS


Bear Problem STEPS

References:

Feed, Feed Storage, & Feeding Practices
Mortality Classification

4.36 Site and staff disinfection and biosecurity

Rationale: All necessary precautions are taken to ensure that pathogens are kept out of a facility. This SOP addresses section *2.2* and *2.3.4.3.3* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that staff members decrease the risk of pathogens being transferred onto the site or between groups of fish.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

4.36.1 ***Personnel Movements:***

Staff should not visit more than one site on the same day. If this is unavoidable, staff members should disinfect footwear between sites, and change into clean, dry clothing if appropriate.

Footbaths and hand wash stations should be used when provided.

If a site has a known disease problem occurring, that site should remain isolated from other sites; the site should be visited only if absolutely necessary and the visitor should not visit any other sites that day or return to his/her normal work site.

Where possible, use of personal protective equipment, including raingear, should be limited to single sites and is to be disinfected after use.

4.36.2 ***Visitors***

As publicly funded hatcheries, we have a unique mandate which includes public education and involvement. Visitors can compromise site biosecurity in a number of ways. They may inadvertently transport pathogens unto a site or may pose a risk to fish and tanks directly with the accidental turn of a valve. Site biosecurity protocols address minimizing the movement of pathogens onto the rearing site and prevention of pathogen movement throughout the site. This SOP addresses section 2.2.3 of the General Principles of Fish Health Management.

Visitors are welcome on our sites during posted business hours. On arrival, visitors should be directed to either contact head office to arrange a guided tour; or where available, self guided tours are available with interpretive stations.

Footbaths and hand wash stations should be placed at critical locations throughout the site and visitors should be expected to use them.

Visitors should be informed not to handle feed, fish or equipment and should be advised to not visit another fish rearing facility within 24 hrs.

Areas holding critical life stages (i.e. incubation rooms) should be off limits, as will any areas holding potentially compromised fish (i.e. broodstock, fish showing signs of illness).

Tours should be conducted following the traffic patterns established to prevent the spread of disease within the facility, i.e. moving from observing the youngest to the oldest fish

All staff and site biosecurity protocols should be followed. (See Site and Staff Disinfection SOP)

All visitors must report to the office; no tours are conducted without staff supervision. Visitors and volunteers will be provided with an orientation but will only be allowed in incubation under direct supervision. All visitors are to be logged in.

4.36.3 ***Supplier Procedures***

Suppliers can transport pathogens from one site to another as they make deliveries and pickups at farm . This SOP addresses section *2.2.5* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that the risk of suppliers moving pathogens onto the site is minimised.

All suppliers should be informed of site procedures and biosecurity protocols prior to delivering anything to the site or removing anything from the site. (Refer to *Site and staff disinfection and biosecurity*)

All deliveries should be made to the front of the main building and/or workshop only. Delivery trucks should not enter the area of the facility where fish are contained, particularly in the event of an outbreak or a suspected outbreak.

It is preferable for suppliers not to visit more than one fish producing site per day. Where this is unavoidable, they must adhere to biosecurity protocols.

When there is a known infectious agent on site, suppliers should be warned in advance, to allow them to modify their delivery schedule to protect other sites on their route. Where possible, the infected site should be the last site visited in the day and the delivery vehicle/vessel should be disinfected after visiting the infected site. When disinfection is not possible, transferring supplies to a site vehicle outside the gates of the site might be a consideration.

Delivery vehicle enter the site and are directed to the unloading location. Staff are responsible for unloading supplies.

4.36.4 ***Facility Maintenance:***

All rearing and holding units, tanks and other containers should be kept clean and tidy.

All floors in fish holding or rearing areas should be kept clear of non-essential equipment, fish food, dead fish, debris, etc.

During incubation and early rearing, footbaths and hand wash stations are placed at critical locations throughout the site, notably the entrance and exit points of the incubation and rearing units.

4.36.5 ***Disinfectant protocols:***

Disinfectants include chemical products determined by the site management in consultation with the Veterinarian.

Products are used according to manufacturer's directions.

Organic matter must be removed from boots and equipment prior to disinfection to ensure efficacy.

Disinfectant concentrations are maintained by visual inspection and by test kits (Virkon) and regular scheduled renewal of the product.

Disinfectants are collected in a small tote and disposed of to ground.

Forms & Records
Visitor log
Volunteer and visitor agreement – waiver sheet

References:
Site and staff disinfection and biosecurity
Equipment disinfection
Chemicals & Disinfectants: Supplies and Storage
Quarantine/Isolation Procedures for Suspected Disease Outbreaks
Outbreak – Disinfection Protocols
Outbreak Response

Egg Disinfection

4.37 Water quality monitoring

Rationale: Maintaining good water quality is vital to good fish health. Monitoring will vary between the sites due to location and site specifics. This SOP addresses section *2.1.1.2* of the General Principles of Fish Health Management. The goal of this SOP is to ensure that water quality is monitored consistently and accurately.

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

General Principles:

Water quality management requires the consideration of fish density, feeding rate, volume, and source supply. If densities or feeding rates are too high, and/or if water volume and/or quality are too low, fish health will suffer significantly.

Tanks are kept clean and water flows are sufficient to maintain dissolved oxygen levels and remove metabolic wastes. Water quality should be measured frequently enough to differentiate normal variation from declining water quality conditions.

Indications for spot-testing water include: losses from an unknown source, temporary rearing at higher than normal densities, behavioural changes associated with water quality compromise (fish gasping at surface or crowding at inflow), historical patterns (i.e. seasonal high water temperatures can be associated with critically low dissolved oxygen), if fish show signs of distress after eating when the metabolic oxygen demand is the highest, etc.

Parameters measured and frequency of those measurements will vary between facilities and their water source and whether water is recirculated or single pass. A water quality monitoring program should be designed to consider natural spatial and temporal variation in water quality and provide an overview of the variation of water quality within a culture facility.

Details of the Operating Procedure:

4.37.1 ***Temperature***

Temperature is measured on a daily basis. Measurements are taken at representative locations throughout the facility. Temperatures are measured using a data logger. Data loggers are placed in incubation, adult holding and juvenile rearing.

4.37.2 ***Dissolved oxygen***

Dissolved oxygen (D.O.) is measured once a week after feeding. D.O. is measured for the inflow and outflow. Ideally D.O. is measured constantly, this is particularly important in recirculation facilities. Measurements are taken at representative locations throughout the freshwater facility e.g. in each tank influent and effluent, in header tanks etc….

The D.O probe is calibrated according to manufacturer's directions prior to use each day. Specifics of probe calibration are to be included in SOP or a link to manufacturer's instructions.

A DDC measures the dissolved oxygen at the outlet of ponds 9-12 and sends the data to the computer located in the main office building. If required, the oxygen levels are measured using an Oxy-Guard handheld meter. This may be carried out during treatments or when flows are reduced.

4.37.3 *Other Parameters*

Flow is metered in the aeration tower and at each well. The water flows are logged to the computer in the main office building.

4.37.4 *Aeration Tower Overflow Chart*

Inches (Head)	mm (Head)	L/Min
1.0	25	400
1.5	38	730
2.0	51	1120
2.5	64	1565
3.0	76	2050
3.5	89	2830
4.0	102	3450
4.5	114	4100
5.0	127	4800

Form & Records:


Pond DO log oct 2007.xls


Water Quality.xls

References:

Water quality contingency plan


Surge Tower.doc

4.38 Water quality contingency plan

Rationale: Acute deteriorations in water quality can result in mass mortality of fish populations. A plan needs to be in place to protect fish from declining water quality. This SOP addresses section *2.1.1.3* of the General Principles of Fish Health Management.

The goal of this SOP is to have a system in place that will protect fish from catastrophic poor water quality events. Examples of catastrophic water quality failures include issues both within (pump failure, pipe burst, filter clogging, etc.) and upstream of the hatchery (turbidity events from landslides, chemical spills during transport, etc.).

Authority: The information contained within this SOP will only be revised by the Fish Health Management Team. Hatchery personnel are responsible for carrying out the procedures contained within this SOP and for ensuring that the SOP is carried out correctly.

Details of the Operating Procedure:

A water quality monitoring program must be in place (see *Water quality monitoring*).

Tanks are equipped with alarms to address water flow rates, water level in header tanks and possibly critical water quality parameters like dissolved oxygen levels. Alarm systems vary with the type of water supply. A designated staff member is on standby to respond to alarms 24/7.

Level alarms (low and high level) are installed in incubation

A site may have access to a variety of water supply options. This redundancy in supply has multiple advantages. It allows the cleanest possible water to be directed to sensitive lifestages (i.e. well water during egg incubation), allows mixing of different water sources (i.e. well water and surface water) for cost-effective temperature manipulations, and allows immunocompetent juvenile fish to be reared in the same water they will eventually be released into, allowing imprinting to their native streams and controlled exposure to endemic pathogens. The main value of redundant water sources however, is that in the event that one water supply is compromised, alternate sources may be available till normal supply is restored.

There are two main water sources available at the Nitinat River hatchery.

- Surface river water is filtered and pumped to adults and older fry. 6 pumps are available for supplying river water.

- Ground (well) water is pumped for use in incubation and for Sarita Chinook, and for fry after clipping and vaccination. 8 ground (well) water pumps are available.

Fish are monitored multiple times daily. A disease outbreak will initially have visible effects on susceptible, individual fish while overall the population may appear normal. However, in the event of a water quality failure, all fish on that water source will be similarly affected.

Backup gas and diesel generators are on site to ensure power is available in the event of a power failure. These are tested to ensure they are functioning properly. Backup pumps are in place and regularly tested.

A disadvantage of flow through water supplies is that in the event of water flow shortage or water quality failure, large numbers of fish in tanks and raceways will quickly exhaust the dissolved oxygen levels in stagnate water. The hatchery has the capacity to provide supplemental oxygen via air stones in an emergency, however this is limited and not available to the entire site. Nitinat has an oxygen generator on site that can supply oxygen directly to the deep ponds.

Where water flows and/or supplemental oxygen capacity is limited, our sites have a last ditch option to do an emergency, early release of progeny from native broodstock, thereby freeing up resources for non-native stocks. This option will only be exercised if the fish in question are have not been showing signs of illness, are not on medication or subject to any medication withdrawal time restrictions.

Coho can be released from the shallow ponds to the river.

A priority release list is maintained and varies with time of year. Generally chum will be released first, then Chinook or coho depending on when the need occurred. In an emergency water will be devoted first to incubation. There is enough redundancy with river water and DH 1 to supply incubation.

If it became an absolutely necessity, fish can be released from anywhere on the site.

If no mitigation options are available, or if unable to prevent fish losses, humane euthanasia of compromised stocks is preferable to suffocation (see *Euthanasia*).

If mitigation efforts are not successful and losses are high, the premises should be quarantined until it is determined that a disease outbreak is not occurring (see *Outbreak Response*

4.38.1 *Emergency Operating Procedure*

Equipment – Generator Set #1 & Operation of the Manual Switches, MS1 & MS2 (located on wall beside the Generator)

These procedures may be required under the following circumstances:

1. ***Failure of the ATS Panel*** for any reason, ATS (Automatic Transfer Switch) is unable to deliver the required voltage and amperage, **and** is unable to operate properly when a manual transfer of this switch is attempted.

    - Possible that switch has arced out and welded its contacts
    - Internal shorting of the main bus bars, Ground fault condition

2. ***Emergency Maintenance to MDC*** (Main Disconnect Centre)

    - Replacement, Repairs, or a need to swap one of the main breakers in MDC
    - MDC has gone into a Ground fault condition

- Disconnect for MCC4 does not work at the MDC – In this case MDC does work and so does ATS & ASP

1. Turn off the Main Breaker on the MCC4 - Panel (located at the River Pumphouse)
2. Turn off the MCC4 Main Disconnect (located at the MDC Panel in Main Powerhouse)
3. Put MS1 & MS2 to the manual operating position (located beside Genset #1)
4. Turn Genset #1 to the off position at the ASP (Automatic Synchronisation Panel)
5. Start the Generator #1 using the Manual switch on the Engine Controller Panel

***Note: If for some reason the engine has to be started by bypassing the Engine Controller**:*

   a. *Go to the small panel that is mounted on the Generator frame*
   b. *Use the jumper switch that is inside the box (ensure it is in the off state) plug it into the centre, of terminals (#1 & #8) Give it a twist to lock it in place.*
   c. *Turn the switch on (this enables RSP, - the fuel solenoid relay to open)*
   d. *Find the solenoid CBX mounted on the opposite side of the generator – Locate the 4 wires on it.*
   e. *The 2 larger wire are for the contacts that deliver 24 volts to the starter solenoid*
   f. *These wires can be jumpered across here, or directly at the starter to start the diesel*
   g. *Once started remove only the wire used to jumper CBX*
   h. *If you need to stop the diesel for any reason the switch jumpered to RSP needs to be turned off, this will cut the fuel supply and stop the diesel*

6. Turn on the Main Breaker on the MCC4 – Panel
7. Start your Well Pumps – DH6, DH7, & DH8

Note: Do Not try to run RDP6 with the 3 above wells, the generator cannot handle the extra load – RDP6 should more than likely already be running if this situation has come this far.

Forms & Records

alarm & callout sheet.pdf

Water Sampling Sheet tag.doc

Standby duties

References:

Outbreak Response

Euthanasia
Water quality monitoring

5 Appendices

5.1 BKD sampling procedure (revised 2006)

Bacterial Kidney Disease Screening Program

Bacterial kidney disease caused by *Renibacterium salmoninarum* (Rs) is a significant cause of mortality in salmonids in enhancement programs in the Pacific Northwest. This disease can cause high mortality rates in hatcheries where there is intensive rearing of fish. In order to reduce the impact of this bacteria on fish in enhancement facilities the diagnostic laboratory has a brood stock screening program to identify fish at high risk of vertically transmitting Rs. Hatcheries that have had a history of BKD in their juvenile fish are encouraged to participate in this screening program. The screening program allows hatchery managers to monitor their BKD status and plan for intervention methods. Once fish are identified as high risk transmitters of Rs a management decision can be taken to reduce the risk of vertical transmission of BKD. By identifying hatcheries with a high percentage of Rs positive brood stock we will be better able to recommend intervention strategies such as erythromycin injection of brood stock prior to spawning or treatment of the high risk progeny.

An ELISA for BKD is used to screen the female brood stock[4] from participating hatcheries. This test measures as low as 20 □g of the Rs antigen (soluble) P57 (Pascho and Mulcahy al., 1987). The kidney from females is used to test for infection since it is the target organ for Rs. Fish that test moderately positive and highly positive on ELISA are re-tested with a direct fluorescent antibody technique (DFAT) Bullock et al., 1980 for confirmation of the results.

The criteria used for identifying positive fish on the ELISA are based on those of Pascho et al., 1987, and Pascho et al, 1991, in which a fish with an ELISA absorbency reading between two standard deviation above the negative control value and 0.199 was considered low, an absorbency reading of 0.2 to 0.999 was considered moderate, and an absorbency reading greater than 1.0 was considered high.

Action Plan

1) Eggs from fish which test negative or low on the ELISA will be pooled and reared using standard fish culture methods.
2) Eggs from fish with a moderate positive ELISA readings will be segregated and reared separately from the eggs in Group 1 (negative or low).
3) Eggs from fish with high absorbency ELISA readings for Rs will likely be discarded. In some cases they may be segregated for separate rearing (from Groups 1 and 2).

Evaluation and determination of appropriate action is made on a case by case basis depending on the stock and the facility in question.

Goals

[4] Rs infected eggs are thought to be a major source of infection for fish reared in captivity (Bullock et al., 1978, Evelyn et al, 1984)

The goals of this screening program are to reduce the likelihood of spreading Rs in hatchery facilities. In addition, this program will enable us to identifying hatcheries which could benefit from brood stock erythromycin injections prior to spawning and as well as groups of fish in the hatchery which may require BKD treatment.

Only females are to be sampled

1. Label Whirl-pak™ bags with a waterproof felt pen.

2. Put the scalpel and tweezers into a beaker of alcohol. Burn off the alcohol by passing the blade of the scalpel and the tweezers through a propane torch flame (called "flaming"). Tools may be laid across the top of the beaker until used.

3. Do an external examination. Record any abnormalities.

4. Pull away swim bladder and other internal organs using the scalpel handle. Start at the anterior (head) end of the swim bladder and pull down and towards the tail end. DO NOT TOUCH the middle or posterior (tail end) kidney with the scalpel handle!

5. Dip the scalpel blade in alcohol and "flame" the blade. Cool for a few seconds.

6. Cut a chunk of kidney (about 1cm wide x 1cm deep x 2 cm long, or roughly the distance between the tip of your thumb and the knuckle) from the posterior portion of the kidney. Use the tweezers to put these into the labeled Whirl-pak bag and seal. DO NOT TOUCH THE KIDNEY SAMPLE WITH ANYTHING BUT THE TWEEZERS!!! Put the Whirl-pak™ bag on ice in either a garbage bag or cooler to keep the samples cool. Wipe the scalpel blade and tweezers with Kleenex and return them to the alcohol beaker. Frequently change the scalpel blade as it becomes dull easily and scrub tweezers with a wire brush to keep them clean.

7. Examine the internal organs and record any abnormalities.

8. Discard any eggs from fish with obvious pustules in the kidney or if the ovarian fluid is cloudy.

9. Phone the lab at 250-756-7057 with the sample size, then ship the samples (with ice packs) the following morning. Samples must reach the lab ASAP after field collection. Please include the address of the hatchery and the phone number of the contact person. If you are shipping by air, we need to know the airlines, arrival time at Nanaimo Airport and the airline's waybill number.

10. It is VERY IMPORTANT that the samples be kept cool at all times. They are to be frozen if shipment must be delayed for more than one day. Please indicate which samples have been frozen and which are fresh.

Required sampling equipment for fall BKD survey

1. Small (6 oz.) Whirl-pak bags ---one per fish
2. Scalpel handle with blades
3. One pair of Tweezers

4. Isopropyl alcohol
5. Small container for alcohol and instruments, preferably with a layer of wax in the bottom
6. Kleenex
7. Propane torch or alcohol burner to sterilize instruments
8. Waterproof felt pen for labeling
9. Container with ice of freezer packs to keep samples cool
10. Garbage bags or other plastic bags to protect samples from the melting ice water

Sources of equipment

Prices are subject to change – use these prices as an approximation.

FISHER SCIENTIFIC 1-800-234-7437

Disposable-Blade Dissecting Knives Size 4 Stainless steel
Cat. No. 08-917-5 Approx. $18.00 each

#22 Stainless-steel Blades
Cat. No. 08-918-5C Approx $45.00 for pack of 100

Blunt-Pointed Forceps---Curved
Cat. No. 08-875-5 Approx $4.00 each

VWR SCIENTIFIC 1-800-932-5000

6 oz. Whirl-Pak Disposable Sampling Bags
Cat. No. 11216-012 Approx $95.00 for box of 500

5.2 Collection of Ovarian Fluid for IHN Virus Assay

Collection of ovarian fluids is carried out during spawning.

Using a sterile transfer pipette, withdraw 2 – 3 mls of ovarian fluid from the container into which the eggs have been spawned. Put the fluid into a sterile, non-breakable, leak proof container. Plastic screw cap test tubes are recommended.

Using a waterproof marker, write the fish number on the container.

The container into which the eggs have been spawned should be cleaned and wiped with paper towel between each fish.

Change pipettes and collect the next sample.

Collection containers (test tubes) must be kept cool at all times.

Containers are put into TWO plastic bags, securely sealed and placed on gel or freezer packs for shipment.

The samples should not be frozen unless shipment is to be delayed for more than 7 days. Keep samples in a refrigerator until shipped.

Keep the shipment cool by using ice or freezer packs.

5.3 Sample submission form

Fish Pathology Laboratory
Pacific Biological Station
Nanaimo, B.C., V9T 6N7
Tel: (250) 756-7057 Fax: (250) 756-7053

DATE: _____ HATCHERY OR SAMPLE SITE:_____

SUBMITTED BY:	MAILING ADDRESS:
_____	_____
PHONE:_____	_____
FAX:_____	_____

SAMPLE INFORMATION

BROODSTOCK CODE: _____ LAB CASE NUMBER:_____

SPECIES:_____ SAMPLE SIZE: _____

SAMPLE TYPE (√):

RANDOM		MORTS		SICK		NORMAL	

REARING CONTAINER I.D.:_____(TROUGH/TANK/POND)

AGE (FROM HATCH): _____AVERAGE WEIGHT (gm):_____

DIET:_____

WATER SOURCE: (√)

SALT		RIVER		LAKE		WELL		SPRING		MIXED		CITY	

TEMPERATURE:_____ºC OXYGEN (AVERAGE):_____ PPM

NUMBER OF FISH IN REARING CONTAINER: _____

LOSS RECORDS (PLEASE INCLUDE DATE):

TODAY:_____PAST 10 DAYS:_____

REASON FOR SUBMISSION:_____

_____

DESCRIPTION OF FISH BEHAVIOUR , APPEARANCE, AND OTHER PERTINENT INFORMATION:

_____

_____

Ovadine™ Emergency Drug Release (EDR) – Hatchery Reporting Requirements

Ovadine is commonly used at fish hatcheries for equipment disinfection. It has also been safely used for over two decades as an egg surface disinfectant during water hardening.

Ovadine is currently (January 2007) under review by the Veterinary Drug Directorate for approval as a fish egg disinfectant. Until approval is received, it is available only by prescription from a licensed veterinarian, through Health Canada's Emergency Drug Release (EDR) program.

The EDR process:

The Veterinarian applies for approval for Site X to use X litres of Ovadine to disinfect X Pacific salmon eggs.

For example: PBS Hatchery - 20 L of Ovadine to disinfect 4 million eggs.

If approved, the site is issued an EDR#, which can be used to order Ovadine from an approved supplier (currently Syndel is the only approved supplier). Along with the EDR#, you will be supplied an Ovadine Usage Record Form (see next page). This form must be filled out when using Ovadine on eggs and must be returned to your Veterinarian within 6 months, to be included in a report to Health Canada. If you see any problems with your eggs or hatch that you think might be related to Ovadine egg disinfection, this form is where you record your concerns. This is an important part of the approval process.

If you need Ovadine only for equipment disinfection, you may order and use it without keeping any records.

If you already have sufficient Ovadine on site for both equipment disinfection and egg disinfection needs, you must still inform the Veterinarian to receive approval for its use on eggs and you are still required to keep records of the amount used on eggs.

If you require Ovadine, you must contact Syndel Laboratories and order the total volume desired for all disinfection needs. To complete the order, you must quote the EDR # for the volume (in litres) for which you have been approved of for egg disinfection.

For example: Order 56 L of Ovadine with 20 L to be used for egg disinfection under EDR # 2006-17xxx.

Use Ovadine as directed for egg disinfection. At the end of each spawning day, record the total volume of Ovadine used on eggs on the following form.

After incubation is complete, or when requested to do so, please return the Ovadine Use Record Form to your Veterinarian.

5.4 Ovadine™ Six Month Usage Record

(To be returned to the Veterinarian at the end of the spawning season)

EDR#: _____

DFO Facility Name: _____

Ovadine supply company: _____

Lot number and expiration: _____

Veterinarian: _____

Each date used on eggs	Volume used on eggs only (L)	User initials	Comments (include species, number of eggs treated and any adverse effects noted)

5.5 Guidelines for in-Stream Placement of Salmon Carcasses for Nutrient Enrichment

Introduction

Historically, large numbers of salmonid carcasses provided entire watersheds with abundant nutrients and organic matter derived from the ocean. Recent research strongly supports the hypothesis that salmon carcasses play a key role in maintaining the productivity of salmonid systems and benefiting the aquatic and terrestrial ecosystem as a whole. Rearing juveniles consume salmon eggs, feed directly on spawned-out carcasses, and benefit from increased abundance of invertebrates and algal growth. The presence of carcasses in streams has been related to increased juvenile density, growth rate, body size, improved fish condition, improved over wintering survival and ultimately increased marine survival.

These guidelines have been developed to regulate the in-stream placement of hatchery salmon carcasses from Fisheries and Oceans Canada enhancement facilities where there is a desire and the capacity to distribute carcasses. The guidelines are not intended to enforce the distribution of carcasses, nor to replace harvest under an Excess Salmon to Spawning Requirements (ESSR) authorization.

These guidelines are meant to increase the overall benefits from carcass placement by minimizing disease risks and other concerns, providing general management strategies for carcass placement, and highlighting the interagency process to avoid conflicts with potentially affected groups and agencies. Numerous factors affect the benefits of carcass placement in streams. These include ambient nutrient content in treatment streams, abundance of native salmon spawners, presence of fish disease agents in carcasses, retention and distribution of carcasses in waterways, water temperatures, flow levels, light penetration, and predator / scavenger activity on carcasses by insects, fish, birds and mammals. These factors have been considered in the development of the guidelines The guidelines were developed utilizing current relevant literature, input from DFO fish health specialists and ecological research scientists, and guidelines prepared by the Washington Department of Fish and Wildlife.

Planning, Review, And Awareness

Carcass placement plans must be reviewed by a DFO member of the Introductions and Transfers committee. Projects that meet the terms of the carcass placement guidelines will be issued a letter from the Department allowing the transport and deposition of carcasses. This letter must accompany all carcass movements. Carcass placement plans should be discussed with all relevant groups and agencies. These groups will include DFO local area staff in stock assessment, habitat, and resource management, and Conservation and Protection (Fishery Officers), as well as local First Nations, stewardship groups, affected landowners or any other affected groups. It is also important to contact the regional Ministry of Environment office to ensure that carcass placement is coordinated with inorganic nutrient enrichment projects. The Ministry of Environment should also be contacted if placement is considered in non-anadromous waters.

Under the Water Act, downstream water users (primarily local municipalities), must be advised of activities that may potentially impact water quality of their withdrawals. Accordingly, Water Licensees on treatment streams should be advised prior to placement programs. Carcasses should be distributed in such a way so as to avoid or minimize impacts on domestic and other types of intakes or water

supplies. Background material and signage may be provided to advise members of the public of carcass placement activity and its benefits.

Carcass Management and Condition

The placement of salmon carcasses in streams may pose a risk of disease transmission if carcasses of infected fish are used, if carcasses are moved to areas within the watershed that are normally not accessible to salmon, or if carcasses are moved to streams outside the watershed. Streams that receive carcasses are referred to as "treatment" streams and those that provide carcasses are referred to as "donor" streams. In general, no carcasses may be moved outside their natal stream because of concerns regarding disease transmission. However, in specific circumstances, movement of carcasses from the watershed to nearby streams may be considered if all of the following conditions are met: donor and treatment streams are geographically proximate and, treatment stream is within the zone of influence of the donor stock (i.e. adults may be straying from donor to treatment stream), and current disease history is available. If sufficient information is not available, health testing of fish in the donor stream and treatment stream may need to be undertaken. Historical information can be obtained by searching the Pacific Biological Station (PBS) Fish Health Database; the Fish Pathology Program may be contacted at (250) 756-7057. Please note that wild fish surveys have not been conducted in many locations in recent years so that information contained in the database does not include current disease status for many salmon stocks.

Only those fish killed with CO2 or blunt trauma that show no visible evidence of serious disease should be used for carcass placement. Carcasses of recently dead salmon from managed spawning channels may also be considered for placement.

Because of drug clearance times, and the length of holding, fish previously treated with an antibiotic or chemical anaesthetic (i.e. TMS™, Aquacalm™) must not be used for carcass placement. However, fish treated with external chemicals that do not require a withdrawal period (e.g. Parasite-S™ or Chloramine-T) are considered safe for placement. If in doubt, contact the Fish Pathology Program. Carcasses may be frozen for later use. However, as freezing will not significantly reduce disease organism loads, it should not be considered a disease management tool.

Carcass Loading Density

All salmonid carcasses are considered equal from a nutrient content basis. That is, required placement load may be calculated as biomass and then converted to fish numbers of the available species. For example, Chinook carcasses may be substituted for coho, and vice versa. Where system-specific weight data are not available, the following average weights for returning B.C. salmon are provided for weight conversion.

<u>Suggested Average Weights for B.C. salmon *</u>
Pink 1.5 kg
Steelhead 4.0 kg
Sockeye 2.5 kg
Chum 4.5 kg
Coho 3.0 kg
Chinook 8.5 kg
* Data sources: mean weights from B.C. catch statistics (J. Bateman, pers. comm.)

The maximum carcass placement within a stream segment (including the areas into which carcasses drift from the distribution point), over the course of a spawning season should be 1.9 kg/m² based on Wipfli et al. (2003) and WDFW (2002). In treatment streams with continuous escapement records, the carcass numbers may be reduced by the recent 10 year average for natural escapement to the treatment reach.

For determining total carcass deposition maximums for streams used by more than one salmon species, the area historically available to each salmon species should be used to calculate the loading rate. Spawning timing should be factored into distribution schedules.

Maximum loading densities may be adjusted to reflect the stream's carcass retention properties. Carcass retention in streams is affected by predator / scavenger activity, carcass transport during high flows, and abundance of in-stream structures to catch and retain carcasses. Accordingly, for streams with expected good carcass retention, maximum carcass densities may be reduced by the current spawner densities. For streams with expected poor carcass retention (high gradient, high flows, few pools and few in-stream structures), carcass loading densities need not be adjusted for current spawner densities.

Carcass Distribution

The temporal and spatial distribution of carcasses should reflect the historic spawn timing and abundance of salmon in the treatment reach. Carcasses should be placed in stream areas that are normally (or recently historically) accessible to salmon, (i.e., not above barriers). Carcass placement into inaccessible stream segments may be permitted where juvenile salmon of the same stock and species have been previously out planted (e.g., colonized upper areas above impassable barriers) but consultation with regional Ministry of Environment staff is necessary.

Placement in the riparian zone is not necessary and often results in increased numbers of blowflies. (Reimchen et al, 2003.). Natural predators will remove carcasses from the treatment stream and distribute them in riparian zones.

For streams with poor access (and low public use), a few accessible sites may be used for regular carcass placement. These sites should be inspected periodically to ensure adequate natural dispersion of carcasses. Where dispersal is poor, carcass loading should be reduced.

Carcasses should be distributed in stable stream areas, where possible. This will help avoid rapid downstream transport of carcasses. Optimal sites include shallow backwater pools, side-channels, small headwater tributaries, areas with abundant woody debris and beaver-dam complexes. However, note that placing excessive numbers of carcasses in side pools with sluggish or intermittent water exchange may cause de-oxygenation (E.A. MacIsaac, pers. comm.). Carcass placement should be avoided or delayed during high flow events, especially where anchoring and/or riparian placement is not feasible. Carcass distribution schedule should consider anticipated problems of poor stream accessibility due to snow, high water, and other constraints.

Timing of carcass placement is also important as nutrients should be made available to young salmon upon their emergence from the gravel. Placement timing may be early, mid or late, and may be used to influence the ecological response to loading within watersheds. For example, the use of carcasses from

later runs of native salmon (fall and winter) may benefit the next growing season, provided that some nutrients are stored through the winter (Wipfli et al. 1999). Also, the use of carcasses from several species, each with a different run timing (e.g., early sockeye, mid-chum, late coho), will provide a longer nutrient pulse in the treatment stream than if only one or two species were used, each with a brief spawning period.

If a treatment stream has a late natural spawning timing, carcasses from earlier runs to the treatment stream may be frozen and stored for later placement. The use of frozen carcasses is also convenient for long-distance transport.

Carcass Anchoring/Mutilation

Carcasses may be tethered or anchored in place, especially in unstable, higher-flow areas in order to improve carcass retention. Where carcass anchoring is desirable, natural anchors (e.g., large woody debris, logjams, beaver-dams) or bio-degradable tethers such as natural-weave ropes, should be used where possible. External identification tags should be removed from carcasses prior to their placement. Non-bio-degradable tethers should be collected and removed from the stream after carcass decomposition. Where frozen carcasses are used, they should be tethered in place (frozen carcasses float and may be readily transported downstream). Where tethering is not possible, it is preferred to thaw out at least one fourth of the frozen carcasses before distributing them in order to enhance carcass retention at the point of access.

Where escapement enumeration programs will be conducted on treatment streams, carcasses should be cut in half or otherwise mutilated at placement, as directed by area stock assessment staff. This is crucial in order to avoid double-counting and ensure that enumeration programs are not affected.

Records of Carcass Placement

Records of numbers and species of carcasses placed in treatment streams should be maintained in annual data summaries, including areas and dates of placement. Summaries should be provided to the contact member of the Introductions and Transfers Committee.

References and Background Literature

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5.6 National Aquatic Animal Health Program

The following excerpt explaining the functions of the NAAHP are extracted from: http://www.dfo-mpo.gc.ca/Aquaculture/health-sante_e.htm

The NAAHP is a science-based regulatory program for aquatic animal diseases which have been designated reportable or notifiable in Canada because of their potential impact on trade and our economy. The program consists of measures needed to prevent, control and/or eradicate aquatic animal diseases of concern. The NAAHP is modeled after Canada's internationally recognized terrestrial animal health program, and will respect the health measures of the Aquatic Animal Health Code of the World Organization for Animal Health (OIE).

The NAAHP is comprised of the following key elements for listed diseases of concern:

- Listing of aquatic animal diseases meeting international and national criteria for mandatory reporting
- Legislation, regulations and policies
- Surveillance (early detection), monitoring and reporting
- Zonation (regionalization)
- Disease databases
- Laboratory diagnostic testing and capacity building
- Quality Assurance/Quality Control
- Scientific research and technology development
- Import controls
- Export certification
- International relationships (influencing setting of standards, trade negotiations)
- Contingency planning
- Disease control and eradication (containment standards and quarantine, disease preparedness and response etc.)
- Education and training
- Risk analysis
- Awareness
- Animal welfare
- Record keeping (tracking and tracing)
- Codes of practice
- Hatchery Program

As a member of the OIE and the World Trade Organization, Canada is obliged to implement OIE standards for trade purposes, including trade in aquatic and terrestrial animals. In addition, Canada is a member of the Food and Agriculture Organization (FAO) and signatory to the Code of Conduct for Responsible Fisheries aimed at conservation of resources for sustainable economic productivity. Canada's major trading partners are adopting regulatory frameworks for their own aquatic animal health programs to meet these international scientific standards. Canada may be required to attest, for export purposes that aquatic animals and their products originate from regions, farms or sites that are free of reportable or notifiable diseases.

The Minister of Agriculture and Agri-Food, who is responsible for the CFIA, and the Minister of Fisheries and Oceans are jointly implementing the federal responsibilities for the NAAHP. This collaboration between Canada's veterinary services and fisheries authority will greatly facilitate Canada's capacity to meet international obligations for aquatic animal health management.

The CFIA provides the overall program lead for the NAAHP under the legislative authority of the Health of Animals Act and Regulations. The Agency is responsible for the disease surveillance/monitoring protocols and control measures for reportable diseases. DFO delivers and oversees the National Aquatic Animal Health Laboratory System (NAAHLS).

Since the management of the wild and aquaculture industries is a shared responsibility in Canada, the NAAHP is designed to respect federal and provincial/territorial jurisdictions. Expertise and collaboration from provinces/territories and industry will continue to be sought to minimize duplication or gaps in an effort to ensure that all aquatic animal diseases are well managed by government and industry.

The Aquatic Animal Health Committee (AAHC) has members which include the Canadian Aquaculture Industry Alliance (CAIA), the Fisheries Council of Canada (FCC), the Aboriginal Aquaculture Association, the Canadian Veterinary Medical Association (CVMA), provincial representatives, Fisheries and Oceans Canada and the Canadian Food Inspection Agency. The AAHC advises the CFIA and DFO on matters relating to the development and implementation of the NAAHP. Information will be shared extensively with all stakeholders as major components of the NAAHP evolve. This approach will ensure a comprehensive and coordinated aquatic animal health management program for Canada.

5.7 COHO FRY SALVAGE GUIDELINES

Introduction

The Area Chief for Fisheries and Oceans Canada, Habitat and Enhancement Branch, South Coast Division, has requested that fry salvage guidelines be developed to provide direction to community groups. A technical committee has been formed, consisting of Kent Simpson (Coho Stock Assessment Biologist), Mel Sheng (HEB Support Biologist), Greg Bonnell (RHQ Assessment Biologist) and Barry Cordocedo (Community Advisor, Nanaimo).

It is common in many streams on Vancouver Island for fish habitat to be de-watered during the summer months, causing salmonid juveniles to be trapped in isolated pools. Significant mortality results from intense predation, crowding, lack of food, temperature stress and, eventually, loss of water altogether.

Public concern for these losses has prompted many individuals to capture stranded fry and relocate them to better habitat. Fry salvaging can have a positive influence on stock restoration if it does not "overload the system". These fry salvage efforts are very rewarding to participants however there is often unforeseen impact on existing wild fry populations. High unfavourable densities can adversely affect growth and survival rates. Thus there is a need for general fry salvage guidelines which will minimize the impact on existing wild fry populations and yet also satisfy public concern over stranded fry.

A. Fry Salvage Plan:

A plan should be developed prior to any fry salvaging operation and should include:

- A list of candidate streams
- A review of existing data on current stock abundance and habitat inventory, including total wetted area during low flow period
- Some level of stream reconnaissance/ monitoring
- Identification of release sites
- A list of participants and their roles
- A list of equipment and vehicles that will be needed
- Notification of appropriate landowners and permission for access
- Notification of DFO and Provincial Fisheries staff, providing contact names and phone numbers
- Acquisition of necessary permits
- Completion of report data requirement
- Documentation of crew safety measures (informing where crew will be, cellular phone/two-way radio, first aid kit, etc.)
- Documentation of crew training (fish ID, first aid, fry collection/transfer procedures, etc.)
- Appointment of a fry salvage co-ordinator.

B. General Guidelines

1. Outplant fry in the same system.
2. Outplant into large areas that remain wetted throughout the summer, not small isolated pools.

3. Attempt to salvage fry when temperatures are below 16 C; use caution above 18 C.
4. Do not plant above barriers if there is no transplant approval.
5. Do not put into pools where high densities of juveniles already exist (>2 fry/m^2).
6. Avoid cobble size habitat.
7. **Do not plant fry into Black Creek**. **In the case of other study streams contact the Community Advisor or Coho Stock Assessment Biologist.**
8. Avoid temperature shock when moving fry: difference in temperature should not exceed 4 degrees C (2 degrees per hour).

Common sense is the general rule

C. Salvage Sites

Community groups will identify which streams dry up, when, where and how many fry are to be salvaged. This will form the beginning of database information. The data reporting sheet includes this information.

D. Release Sites

1) Plant into known accessible barren areas.
2) Plant into large areas of high quality habitat that remains wetted during summer low flow period.
3) Plant into lake where salvaged fry are from lake tributaries or within anadromous range.
4) Transfer fry to interim locations:
   a) Ponds, channels
   b) Hatchery (low priority); retain to smolt stage*
   c) Lake pens (low priority)*
5) Plant above barrier (colonization) **only if transplant approval is in place**.

* Note: When fry are retained and reared at hatchery or lake pens their releases are to follow current outplanting guidelines.

E. STOCKING DENSITIES INTO BARREN AREAS:

A. Stream plants: 2.0 fry/m^2 < 2% stream gradient
0.2 fry/m^2 2% to 5% stream gradient
B. Lake plants: 2500/ hectare (of water less than 10 ft. deep)

6) F. STOCKING DENSITIES INTO UNDERUTILIZED AREAS:

A. < 2% Stream gradient 1.0 fry/m^2

B. 2-5 % Stream gradient 0.1 fry/m^2

Note: With hatchery releases, densities can be higher during winter. Possible release locations are off-channel sites, favourable upstream habitat or heavy weed growth areas in lakes.

G. TRANSPORT CRITERIA:

A. Aeration	0.1 kg/litre	2- 3 hrs
B. No Aeration	0.013 kg/litre	1 hr

H. REPORTING REQUIREMENTS:

- Keep format as simple as possible
- Information should include:
- date,
- exact donor and release sites,
- no.'s,
- stream name/code,
- est. ave. fork length
- Include map showing salvage and release sites
- Accuracy: numbers can be plus or minus 20%
- Salvage and release location: something understandable/ identifiable
- Send reports to appropriate DFO and Provincial Fisheries staff
- Send data initially to HEB Assessment RHQ

I. APPROVALS:

Community Advisors are to submit/renew/update an outline each year to the HEB Area Chief (Bruce MacDonald) identifying what groups/individuals are involved/phone numbers and name of stream(s) to be salvaged. All South Coast area CA's will attached this information to an Annual Licence/Permit which will be forwarded to Province. If there are no changes in the following year, only the date will have to be changed on the subsequent annual licence/permit.
Process will be same as transplant applications renewal.

J. ASSESSMENT:

HEB staff (Mel Sheng and Greg Bonnell) felt that any assessment program would be difficult and costly to do and thought at this particular time SEP biostandards could be applied to predict survivals and productivity of salvaged fry.

K. CONTACTS/NOTIFICATION:

The following agency offices should be notified of proposed fry salvage activities:

- DFO Fisheries Management
- DFO Conservation and Protection
- DFO Habitat Staff
- Provincial Fisheries staff
- This information will be outlined in annual permit renewal.

Letters of notification should include information on groups involved and phone numbers. (See attachment).

5.8 'PRIMARY' FISH PRODUCTION STANDBY DUTIES

Updated Jan 14 / 2002

General duties and responsibilities:

- Designated "Primary" staff, must respond to all standby 'call out' situations and alarms.
- All alarms need to be canceled at the main office alarm panel. Determine "specific" alarm area and initiate the "4 **R**'s". **R**espond to the alarm area. **R**esolve the problem (call out other staff if there is any safety or security risk associated with the resolution of the problem). **R**eset the panel(s). **R**ecord the problem and any response required in the standby logbook.
- Ensure you have a complete understanding of all hatchery biological operations, facility operating procedures and program priorities. This includes: hatchery water source and supply, system plumbing and hydraulics, minimum water flow requirements for all operating containers, rationalization of well and river water during flow interruption, priority emergency release actions, thermal marking and disease issues for all containers, any abnormal fish behaviours and an operational understanding of all mechanical systems including generators and water-pumping equipment required to maintain optimum culture conditions.
- Ensure all visitors have left the site and that all "checked-in" over night personnel are accounted for and orientated to the residence and specific emergency procedures.
- Ensure that you are familiar with and have a good understanding of all local native and non-native community partners, contractors working with DFO area programs, and other government sectors or agencies (RCMP, CO's. C&P, StAD, and NGO partners) that may require use of the hatchery facilities or require support from hatchery employees.
- Ensure you are familiar with all Emergency Procedures including the Business Resumption Plan, Site Occupation Contingency Plan, Fire Plan, Facility Standard Operating Procedures, Nitinat Hatchery Working Alone Policy and "Check-in" Procedure.
- Act as the Senior DFO representative and be able to respond appropriately to ensure the safety and security of all fish, the facility and the people associated with the hatchery.

Walk About and Lockup duties – ***all areas***

- Ensure that proper heat and lighting levels are observed. Turn off all non-essential lighting and fans in all areas except for both powerhouses where interior lighting must be left on and in the DH#I powerhouse the fan must be left on to vent battery gases.
- Turn off any non-required pressurized wash down or compressed air sources.
- Check for any health and safety hazards. Rectify hazard if possible and document.
- Ensure that all fish culture tools and equipment are stored safely and in their proper place.
- Secure all windows and lock all doors in the office and incubation buildings.

Specific duties for "Primary" Fish Production Standby

Office building:

- Turn off coffee machine in lunchroom. Put dirty dishes in the sink and make coffee in AM.
- Check pump status board. Confirm operation of pumps and verify flow requirements.
- Check answering machine regularly and forward messages appropriately.

Incubation Room and Wet lab:

- Check to ensure alarm panel is clear and regularly "spot check" that float alarms are set and functioning properly.
- Visually check that Head Tank water levels are at their "preset inflow mark"
- Visually check discharge flows in all incubation containers. Screens in bulk boxes must be checked and cleaned regularly. Check and clean any clogged river head tank inflow 'socks'.

- Check that the Temp printer is operating properly and the lab heater is working properly.
- Ensure there is adequate river and well flow in the temperature probe sump reservoir.
- Ensure that all “in use” equipment (chiller, marking and egg picking machines etc) are in standby mode or stored in the wet lab after use.

Keeper Channels:

- Visually check inflow levels in all active keepers channels and keeper boxes.
- Clean all keeper channel and keeper box screens regularly. This is required especially during hatch and flood events.
- Ensure all entrance door and outflow black outs curtains are in place.
- Check for signs of predators in the buildings.
- Observe, record and report all abnormal or unusual physical or biological changes such as low flows, excessive silt build up, air bubbles in keeper boxes, alevin movement (smothering, leakage etc.) or signs of predators. Note any changes or adjustments in standby log.

Pond Areas:

- Check inflows and outflows for fish leakage, proper swim-in or release conditions, unusual fish behavior, signs of predators or any other unusual biological or physical changes.
- Confirm piping, nets or covers are winterized or secure during storm conditions.
- **Residence:**
- Ensure all guests have been assigned rooms, orientated to the residence and follow all residence rules. Ensure dinners are being coordinated with people staying in residence.
- Act as the fire Warden in event of a fire. Close all storage, laundry and bedrooms doors.
- Ensure stove, and all small appliances (toaster, coffeepot) are off after use.

Summer standby duties : Follow Working Alone Policy and all Maintenance Standby Duties.