**U.S. FISH AND WILDLIFE SERVICE DRANSIT EAL SHEET**

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<tr>
<th>PART</th>
<th>SUBJECT</th>
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<tr>
<td>713 FW 1-5</td>
<td>Fisheries&lt;br&gt;Aquatic Animal Health Policy Overview&lt;br&gt;Aquatic Animal Health Operations&lt;br&gt;Exotic Aquatic Animal Disease Eradication Plan&lt;br&gt;Aquatic Animal Health Assistance to Non-Service Entities&lt;br&gt;Special Case Aquatic Animal Movements and Controlled Propagation Programs</td>
<td>440</td>
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**ORIGINATING OFFICE**
Division of the National Fish Hatchery System

**DATE**
March 3, 2004

**EXPLANATION OF MATERIAL TRANSMITTED:**
These chapters are being reissued to revise the name of the U.S. Fish and Wildlife Service Handbook of Aquatic Animal Health Procedures and Protocols.

**DIRECTOR**

**FILING INSTRUCTIONS:**

Remove:

- 713 FW 1, 11/25/03, FWM 433
- 713 FW 2, 11/25/03, FWM 433
- 713 FW 3, 11/25/03, FWM 433
- 713 FW 4, 11/25/03, FWM 433
- 713 FW 5, 11/25/03, FWM 433

Insert:

- 713 FW 1, 03/03/04, FWM 440
- 713 FW 2, 03/03/04, FWM 440
- 713 FW 3, 03/03/04, FWM 440
- 713 FW 4, 03/03/04, FWM 440
- 713 FW 5, 03/03/04, FWM 440
Chapter 2  Aquatic Animal Health Operations

2.1 What is the purpose of this chapter? This chapter describes managerial and technical responsibilities, provides systems to monitor the range and distribution of selected aquatic animal pathogens, establishes minimum standards for inspection and monitoring activities, and delineates required aquatic animal health management and reporting activities.

2.2 What are the conditions, criteria, and general instructions for aquatic animal health surveillance?

A. Inspections at Service Facilities. Aquatic animal health officials will complete Service facility inspections, which will remain valid no longer than 1 year from the time the aquatic animal health official signs the inspection report. Service aquatic animal health officials must conduct sampling on a lot-by-lot basis with samples from each lot distinctively marked, maintained, and processed separately. Any time a listed pathogen is detected by inspections, monitoring, or diagnostics, the Service aquatic animal health official must report it on FWS Form 3-226.

B. Special Case Inspection Criteria. Standard inspection procedures may not be possible or appropriate in cases such as: threatened and endangered species, small populations (less than 100) and nontraditional aquatic animals for which there is limited diagnostic expertise. Sample sizes, tissue type (e.g., non-lethal samples or sentinel animals) and laboratory methodologies are at the discretion of the Fish Health Center Director in consultation with appropriate involved entities and consistent with Risk Assessment Worksheet (713 FW 5).

C. General Instructions for Inspection Sampling.

(1) Selection of animals for samples. Sampling will target moribund animals. An unbiased “grab” sample from the rearing unit must make up the remainder of the samples. If the lot of interest is reared in multiple units, sampling must occur in as many units as is possible. Aquatic animals must be alive when collected or, if killed, held on ice for no more than 1 hour prior to tissue removal. Collect all tissue samples in a manner that avoids contamination between aquatic animal lots or from external sources.

(2) Sampling Schemes. Conduct all sampling as per Exhibit 1, unless otherwise specified within this chapter, and as per the U.S. Fish and Wildlife Service Handbook of Aquatic Animal Health Procedures and Protocols (Handbook).

(3) Determining sample size. From an animal population of 10,000 or greater, for which you would like to be 95 percent confident that the sample will include at least one infected individual, the number of animals to sample (rounded to the closest five animals) will depend on the assumed prevalence of infection within the population or the prevalence level that you would like to detect. For a prevalence level of 2 percent collect 150 animals, for 5 percent prevalence collect 60 animals, and for 10 percent prevalence collect 30 animals. In situations where a higher level of collection confidence (e.g., 99 percent) is required or where lethal sampling must be minimal (e.g., species of concern, captive broodstock) refer to the Handbook. The Handbook provides more precise sample sizes (i.e., not rounded to the closest five animals), and provides sample sizes for other population sizes and other confidence levels. Additionally, the Handbook provides instruction on how to further fine-tune sampling based on efficiency of detection techniques.

(4) Minimal sampling criteria for all Service activities. All Service activities will also be conducted in compliance with applicable Federal, State, tribal, interjurisdictional, and foreign aquatic animal health protection guidelines and/or regulations.

2.3 How are samples collected for surveillance processed? Conduct all sample processing as per procedures in the Handbook.

2.4 What additional restrictions pertain to the transfer or handling of gametes, fertilized eggs, or aquatic animals?

A. Compliance with inspections and monitoring procedures. You may not transfer or release gametes, fertilized eggs, or aquatic animals from Service facilities until you complete all pathogen inspection or monitoring procedures as described, or referred to, in this policy. When gametes, fertilized eggs, or aquatic animals from free-ranging (wild) stocks are brought into a facility, they should be isolated from production eggs, aquatic animals, or broodstock until evaluated for pathogen infection (see 713 FW 5).

B. Compliance with regulations or policy. Service aquatic animal culturing, fishery management, fishery study activities, and the shipping of gametes, fertilized eggs, or aquatic animals will be undertaken in compliance with interjurisdictional, Federal, State, and foreign aquatic animal health regulations and guidelines, as well as this policy.

2.5 What additional restrictions pertain to aquatic animal health-related activities at Service primary broodstock facilities?

A. Additional requirements are placed on primary broodstock facilities to further minimize possibilities for spreading listed pathogens into facilities, stocks, and geographic areas previously free of such pathogens. This policy applies to hatcheries or facilities shipping gametes...
interregionally (i.e., primary broodstock facilities as defined in 713 FW 1). As these primary broodstock facilities supply the entire National Fish Hatchery System, and, in many cases, non-Federal hatcheries, it is imperative that the health status for these broodstock be maintained at the Class A level. Known deviation from this policy must be evaluated on a case-by-case basis and approved by the Director.

**B. Sampling.** Broodstock of the same species consisting of more than 1-year-class held in a common water supply may be considered as one lot for sampling purposes; however, sampling of each year-class is recommended. To facilitate the collection of appropriate samples (see paragraph 2.2, Exhibit 1, and the Handbook), the inspecting aquatic animal health official should coordinate broodstock inspections with the spawning season.

**C. Acquisition, Transfer, and Release of Aquatic Animals.** The goal of primary broodstock facilities, relative to the health of aquatic animals, is to maintain their status as Class A. Managers of these stations must take special precautions to avoid the introduction of pathogens listed in these guidelines into all Service broodstock facilities. Do not take gametes from clinically diseased aquatic animals unless a specific program, such as endangered species, requires the use of these eggs. All salmonid fertilized eggs transferred to Service broodstock facilities must be water hardened in a polyvinylpyrrolidone iodine disinfectant (see paragraph 2.7).

**2.6 What additional restrictions pertain to aquatic animal health-related activities at other Service facilities?** All other Service facilities will be managed to maintain the highest pathogen designation code practical, within the requirements and conditions of the particular resource management needs served by each facility. Gametes, fertilized eggs, or aquatic animals may be transferred to these facilities from stocks, populations, or facilities not determined to be Class A, subject to the following conditions:

**A.** Gametes, fertilized eggs, or aquatic animals may be transferred to Service facilities from any source, if the transfer will not change the pathogen designation code or pose a health risk to the animals currently being held at the receiving facility.

**B.** All gametes, fertilized eggs, or aquatic animals transferred to Service facilities from stocks or facilities not designated as Class A, should be held in isolation from other production stocks until their pathogen status can be determined.

**2.7 What specific requirements pertain to the water-hardening and disinfection of salmonid eggs?** Service personnel will disinfect salmonid eggs with a polyvinylpyrrolidone iodine compound, a nonselective germicide used widely in general disinfection of laboratory and veterinary facilities. This compound is approximately 1-percent iodine. The objective of disinfection is to expose the bacteria and viruses to active iodine. Service personnel should read the label before processing.

**A. Use of Polyvinylpyrrolidone Iodine During Water Hardening.** All salmonid eggs must be disinfected during the water-hardening process. Refer to the Handbook for species-specific methods.

**B. Disinfection.** Eggs received at Service facilities from outside sources will be disinfected before they are allowed to come into contact with water used for aquatic animal culture purposes, rearing units, or equipment at the receiving station.
### AQUATIC ANIMAL INSPECTION SAMPLING SCHEMES (MINIMUM REQUIREMENTS) AND REQUIRED TISSUE TYPE FOR ESTIMATING PATHOGEN PREVALENCE

<table>
<thead>
<tr>
<th>Population</th>
<th>Action Involving Population</th>
<th>Viruses</th>
<th><em>Renibacterium salmoninarum</em> (salmonids only)</th>
<th>Other Cultured Bacteria</th>
<th><em>Myxobolus cerebralis</em> (salmonids only)</th>
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<tbody>
<tr>
<td>captive or feral broodstock</td>
<td>disinfected egg transfers</td>
<td>2% POI ovarian fluid and/or kidney/spleen&lt;sup&gt;5&lt;/sup&gt;</td>
<td>5% POI ovarian fluid and/or kidney/spleen&lt;sup&gt;5&lt;/sup&gt;</td>
<td>optional</td>
<td>N/A&lt;sup&gt;2&lt;/sup&gt;</td>
</tr>
<tr>
<td>primary broodstock</td>
<td>disinfected egg transfers</td>
<td>2% POI ovarian fluid and/or kidney/spleen&lt;sup&gt;5&lt;/sup&gt;</td>
<td>5% POI ovarian fluid and/or kidney/spleen&lt;sup&gt;5&lt;/sup&gt;</td>
<td>10% POI kidney</td>
<td>N/A</td>
</tr>
<tr>
<td>live fish</td>
<td>fish transfer</td>
<td>5% POI kidney/spleen</td>
<td>5% POI kidney/spleen</td>
<td>10% POI kidney</td>
<td>5% POI gill &amp; cranium&lt;sup&gt;3&lt;/sup&gt;</td>
</tr>
<tr>
<td>special cases&lt;sup&gt;4&lt;/sup&gt;</td>
<td>sample number, type &amp; methods at discretion of Fish Health Center Biologist in charge</td>
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**Footnotes:**

1. See Handbook for laboratory methods. Actual sample size depends on population size from which samples are being collected, the desired prevalence of infection (POI) to be detected within that population and the desired statistical confidence interval (CI) of the findings. An approximation of such sample schemes are provided in 713 FW 2.2C(3). A table (found in the Handbook), which supplements the above table and 713 FW 2.2C., was extracted from Simon & Schill (1984). This supplemental table presents guidance for more precise minimum sample sizes to be collected for a given population size, POI and CI.

2. N/A = Not applicable

3. Sample most susceptible species on a given water source for entire facility inspection.

4. Special cases include, but are not limited to: warm and cool water species, threatened and endangered species, species of concern, native fish species, animals for which there are no commonly accepted diagnostic techniques, and low captive population sizes (< 100 animals). Objectives for sampling program would be based on a risk assessment and coordinated with involved Service personnel and cooperators.

5. To achieve the necessary sample size relative to the desired POI, the composite sample may comprise any combination of ovarian fluid and kidney/spleen (e.g., ovarian fluid samples from 120 fish plus individual kidney/spleen samples from 30 different fish).