

Method of Sampling

Sending of live fish samples

1. Submit 3-5 weak fishes showing disease signs.
2. Obtain 3-5 normal and healthy fishes.
3. Pack separately the normal from the diseased samples.
4. Live specimen should be transported in water-filled plastic bags with sufficient aeration/oxygen.
5. Water volume should be 20 parts water to 1 part fish.
6. Use double bags to prevent leakage.
7. Place bags in an insulated or styrofoam box, when transporting
8. Put ice packed in double plastic bags to maintain cool temperature in the box

NOTE:

Do not add too much ice that would make the water very cold for the fish. Make sure that specimens arrive to the laboratory live.

Sending of iced fish samples

1. Obtain 3-5 diseased and 3-5 normal/healthy fishes.
2. Wrap fishes individually.
3. Separate normal from diseased fishes
4. Place packed samples in between layers of ice in a styrofoam box or other insulated containers.

NOTE: Iced samples should arrive to the laboratory for analysis within 24 hours.

Sending of fixed fish samples for Histopathology

(If samples cannot be delivered live within 24 hours)

1. Prepare 10% buffered formalin solution.

For 1 Liter 10% buffered formalin solution

100 mL formalin

900 mL distilled water

4 grams acid sodium phosphate, monohydrate

6.5 grams anhydrous disodium phosphate

2. Obtain 3-5 diseased and 3-5 normal/healthy fishes.
3. Kill the fish by overdosing with MS 222 (tricaine methane sulfonate at 1g/500 ml water) or 600 ppm phenoxyethanol.
4. Open the abdomen of large fish for its entire length with one continuous cut.
5. Immerse specimens in fixative at a ratio of 1 part fish tissue to 10 parts fixative (example: 100 g fish : 1,000 ml formalin solution).
6. For large fishes, cut fishes into several parts.
7. Make sure containers are properly sealed before sending to prevent leakage.

Sending of fixed fish samples for PCR Analysis

(For viral detection)

1. Disinfect dissecting tray by wiping with 70% alcohol.
2. Sterilize clean forceps and scissors by flaming.
3. Dissect organs such as brain and eyes (for VNN detection) and fix in 95% or 70% ethanol.

