Manual of Diagnostic Tests for Aquatic Animals

‘Aquatic Manual’
OIE international standards

Terrestrial Animal Health Code - mammals, birds and bees

Aquatic Animal Health Code - amphibians, crustaceans, fish and molluscs

Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

Manual of Diagnostic Tests for Aquatic Animals
‘Aquatic Manual’


Available on-line [www.oie.int](http://www.oie.int)
Purpose of Aquatic Manual

- Provide internationally agreed standardised approach to the diagnosis of OIE-listed diseases (Aquatic Code)

- Facilitate international trade in aquatic animals and their products by:
  - ensuring harmonisation of diagnostic testing
  - avoiding differences in interpretation of results
  - ensuring quality of diagnostic tests

Recognised as the international standard by the WTO
Purpose of Aquatic Manual

- A key and unique document describing diagnostic methods that can be applied to the OIE-listed diseases in aquatic animal health laboratories all over the world ➔ improved aquatic animal health worldwide;

- Describes diagnostic laboratory methods which are suitable for the detection of disease as part of a national aquatic animal health surveillance/control programme, or as part of a programme to underpin claims of freedom from a specific disease;

- To assist with the development of surveillance methodologies for OIE-listed diseases;

- Surveillance programmes aim to determine, from the results provided by standardised lab. methods performed with samples collected according to define rules (Aquatic Code), the health status for a country, zone or compartment for a specified disease.
Who uses the Aquatic Manual?

- Laboratories carrying out veterinary diagnostic tests and surveillance;
- Competent Authorities in Member Countries.
Aquatic Manual

Note:

- Chapter on *Aquatic animal health surveillance* (1.4.) is in the *Aquatic Code*.

  - additional text on surveillance
Part 1 General Provisions
Section 1.1 Introductory Chapters

Ch 1.1.1 Quality management in veterinary testing laboratories

Ch 1.1.2 Principles and methods of validation of diagnostic assays for infectious diseases

Ch 1.1.3 Methods for disinfection of aquaculture establishments
CONTENTS

Part 2

- Recommendations Applicable to Specific Diseases

Diseases of Amphibians – in preparation
Diseases of Crustaceans
Diseases of Fish
Diseases of Molluscs
General information

Each Section starts with general information - amphibians (pending), crustaceans, fish, molluscs

A. Sampling
1. Assessing the health status of the epidemiological unit
2. General processing of samples

B. Material and biological products required for the isolation and identification of pathogens
1. Scope
“For the purpose of this chapter, DISEASE NAME is considered to be INFECTION WITH PATHOGEN NAME.”

2. Disease information

2.1. Agent factors
2.1.1. Aetiological agent, agent strains
2.1.2. Survival outside the host
2.1.3. Stability of the agent (effective inactivation methods)
2.1.4. Life cycle (if applicable)
2.2. Host factors
   2.2.1. Susceptible host species
   2.2.2. Susceptible stages of the host
   2.2.3. Species or sub-population predilection (probability of detection)
   2.2.4. Target organs and infected tissue
   2.2.5. Persistent infection with lifelong carriers
   2.2.6. Vectors
   2.2.7. Known or suspected wild aquatic animal carriers

2.3. Disease pattern
   2.3.1. Transmission mechanisms
   2.3.2. Prevalence (in wild and farmed populations for the detection method used, under different conditions)
   2.3.3. Geographical distribution
   2.3.4. Mortality and morbidity
   2.3.5. Environmental factors (e.g. temperature, salinity, season, etc.)
2.4. Control and prevention

2.4.1. Vaccination
2.4.2. Chemotherapy
2.4.3. Immunostimulation
2.4.4. Resistance breeding
2.4.5. Restocking with resistant species
2.4.6. Blocking agents
2.4.7. Disinfection of eggs and larvae
2.4.8. General husbandry practices
3. Sampling

3.1. Selection of individual specimens
3.2. Preservation of samples for submission
3.3. Pooling of samples
3.4. Best organs or tissues
3.5. Samples/tissues that are not suitable (i.e. not possible to detect)
4. Diagnostic methods

4.1. Field diagnostic methods
(observation of the animal and its environment)
   4.1.1. Clinical signs
   4.1.2. Behavioural changes

4.2. Clinical methods
(effects of the pathological agent on the host, rather than on agent detection)
   4.2.1. Gross pathology
   4.2.2. Clinical chemistry
   4.2.3. Microscopic pathology
   4.2.4. Wet mounts
   4.2.5. Smears
   4.2.6. Fixed sections
   4.2.7. Electron microscopy/cytopathology

4.3. Agent detection and identification methods
(methods that detect, possibly isolate and amplify, and identify the agent)
   4.3.1. Direct detection methods
      4.3.1.1. Microscopic methods
      4.3.1.2. Agent isolation and identification
   4.3.2. Serological methods
5. Rating of tests against purpose of use

- This information is used to determine which test is appropriate for what purpose.

- E.g. a particular method may be highly suitable to diagnose clinical cases of disease in individual animals of a certain age group, but the same method may be unsuitable for assessing the infection status of large numbers of clinically healthy animals.

- Each *Manual* disease chapter includes a Table comparing different methods for targeted surveillance and diagnosis of Disease X.

- It is an assessment of the test’s ‘fitness for purpose’.
<table>
<thead>
<tr>
<th>Method</th>
<th>Targeted surveillance</th>
<th>Presumptive diagnosis</th>
<th>Confirmatory diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Larvae</td>
<td>PLs</td>
<td>Juveniles</td>
</tr>
<tr>
<td>Larvae</td>
<td>d</td>
<td>d</td>
<td>c</td>
</tr>
<tr>
<td>PLs</td>
<td>d</td>
<td>d</td>
<td>d</td>
</tr>
<tr>
<td>Juveniles</td>
<td>d</td>
<td>d</td>
<td>c</td>
</tr>
<tr>
<td>Adults</td>
<td>d</td>
<td>d</td>
<td>c</td>
</tr>
<tr>
<td>Gross signs</td>
<td>d</td>
<td>d</td>
<td>c</td>
</tr>
<tr>
<td>Bioassay</td>
<td>d</td>
<td>d</td>
<td>d</td>
</tr>
<tr>
<td>Direct LM</td>
<td>d</td>
<td>d</td>
<td>c</td>
</tr>
<tr>
<td>Histopathology</td>
<td>d</td>
<td>c</td>
<td>c</td>
</tr>
<tr>
<td>Transmission EM</td>
<td>d</td>
<td>d</td>
<td>d</td>
</tr>
<tr>
<td>Antibody-based assays</td>
<td>d</td>
<td>d</td>
<td>c</td>
</tr>
<tr>
<td>DNA Probes <strong>in situ</strong></td>
<td>d</td>
<td>d</td>
<td>c</td>
</tr>
<tr>
<td>PCR</td>
<td>d</td>
<td>b</td>
<td>a</td>
</tr>
<tr>
<td>Sequence</td>
<td>d</td>
<td>d</td>
<td>d</td>
</tr>
</tbody>
</table>

**a** = the method is the **recommended method** for reasons of availability, utility, & diagnostic specificity & sensitivity;

**b** = the method is a **standard method** with good diagnostic sensitivity & specificity;

**c** = the method has **application in some situations**, but cost, accuracy, or other factors severely limits its application;

**d** = the method is presently **not recommended** for this purpose.
The rating of the different diagnostic methods are somewhat subjective as suitability involves issues of reliability, sensitivity, specificity and utility.

Although not all of the tests listed as category a (the recommended method) or category b (a standard method) have undergone formal standardisation and validation, their routine nature and the fact that they have been used widely without dubious results, makes them acceptable.
6. Test(s) recommended for targeted surveillance to declare freedom from Disease X.

Describes methods, based on the information provided in point 1-4, and assessed in 5, for targeted surveillance to declare freedom from disease as outlined in the *Aquatic Code*. 
7. Corroborative diagnostic criteria

7.1. Definition of suspect case
7.2. Definition of confirmed case

Defines what constitutes a suspect case of disease, and a confirmed case of disease

For example, a certain level of mortality at the right time of the year, in susceptible animals, together with matching clinical signs, liver lesions and histopathology could be sufficient for suspicion of Disease X. Several combinations may be possible.

A confirmed case could be defined where in addition to the above, the agent has been detected. However, detection of viable agents without any clinical signs could also constitute a confirmed case.

This information is required:
- for the purpose of disease investigations, especially in cases where ‘free’ status is threatened.
- when surveillance of healthy populations yields controversial results, e.g. positive PCR signals in the absence of any other evidence of infection.
CONTENTS

Part 3
OIE Expertise

List of OIE Reference Laboratories and Collaborating Centres for diseases of amphibians, crustaceans, fish and molluscs
Reference Laboratories, Collaborating Centres and Twinning Laboratories
OIE Reference Laboratories

- designated to pursue all the scientific problems relating to a specific disease(s) on the OIE list and other diseases or topics of importance for international trade;

- function as a centre of expertise and standardisation of diagnostic techniques for the specified disease(s);

- provide scientific and technical assistance on surveillance/control of the specified disease(s).
Principal mandate:

- standardise the diagnostic techniques;
- store and distribute to national labs. biological reference products/reagents used in the diagnosis/control of the designated disease(s);
- develop new procedures for diagnosis/control of the designated disease(s);
- gather, analyse and disseminate epizootiological data;
- place expert consultants at the disposal of the OIE.
May also contribute to:

- provide scientific/technical training for OIE Members;
- provide diagnostic testing facilities to OIE Members;
- organise scientific meetings on behalf of the OIE;
- coordinate scientific studies;
- publish and dissemination information that may be useful to OIE Members.
World Distribution of the OIE-Reference Laboratories

- 187 OIE Ref. Labs (31 aquatic)
- 36 Countries
- 100 Diseases (25 aquatic)
- 161 experts
Aquatic OIE Reference Laboratories

There are currently 31 OIE Reference Laboratories for OIE listed aquatic animal diseases (25/26 listed diseases)

- each Lab. is under the guidance of an expert whose competence is recognised internationally
- expert is approved by the OIE World Assembly of Delegates
- expert must be a leading and active researcher
- expert must submit an Annual Report to the OIE
- RL must notify the OIE if the expert leaves

If the RL confirms a positive for OIE listed diseases, the RL should immediately inform the OIE Delegate of the Member as well as the OIE Headquarters
Aquatic OIE Reference Laboratories

Contact details of RL listed in the Aquatic Manual

LISTED DISEASES OF AMPHIBIANS
- Infection with *Batrachochytrium dendrobatidis*: Australia
- Infection with ranavirus: Australia (x2)

LISTED DISEASES OF CRUSTACEANS
- Crayfish plague (*Aphanomyces astaci*): United Kingdom, Finland
- Infectious hypodermal and haematopoietic necrosis; USA
- Infectious myonecrosis; USA
- Taura syndrome; USA
- White spot disease: USA, Chinese Taipei
- Necrotising hepatopancreatitis: None - listing adopted at GS May 2010
- White tail disease: India
- Yellow head disease: Australia
LISTED DISEASES OF FISH
- Epizootic haematopoietic necrosis: Australia (x2)
- Infectious haematopoietic necrosis: USA
- Epizootic ulcerative syndrome: Thailand
- Gyrodactylosis (Gyrodactylus salaris): Norway
- Infectious salmon anaemia: Norway, Canada
- Koi herpesvirus disease: Japan, United Kingdom
- Red sea bream iridoviral disease: Japan
- Spring viraemia of carp: United Kingdom
- Viral haemorrhagic septicaemia: Denmark

LISTED DISEASES OF MOLLUSCS
- Infection with abalone herpes-like virus: Chinese Taipei
- Infection with Bonamia exitiosa: France
- Infection with Bonamia ostreae: France
- Infection with Marteilia refringens: France
- Infection with Perkinsus marinus: USA
- Infection with Perkinsus olseni: USA
- Infection with Xenohaliotis californiensis: USA
DELISTED DISEASES OF CRUSTACEANS
- Spherical baculovirosis (*Penaeus monodon*-type baculovirus): USA, Chinese Taipei
- Tetrahedral baculovirosis (*Baculovirus penaei*): USA

DELISTED DISEASES OF FISH
- Bacterial kidney disease (*Renibacterium salmoninarum*): USA
- Channel catfish virus disease: USA
- Enteric septicaemia of catfish (*Edwardsiella ictaluri*): USA
- *Oncorhynchus masou* virus disease: Japan
- Viral encephalopathy and retinopathy: Italy, Japan

DELISTED DISEASES OF MOLLUSCS
- Infection with *Bonamia roughleyi*: France
- Infection with *Martelia sydneyi*: France
- Infection with *Haplosporidium costale*: USA
- Infection with *Haplosporidium nelsoni*: USA
OIE Collaborating Centres

- centre of expertise in a specific designated sphere of competence relating to general matters on animal health (epidemiology, risk analysis, training of official vets, etc.),

- in its designated field of competence, they must provide their expertise internationally

- CC is approved by the OIE World Assembly of Delegates
World Distribution of the OIE- Collaborating Centres

- 35 Collaborating Centres (1 aquatic),
- 20 Countries,
- 33 Topics,
- 35 experts,
OIE Collaborating Centres

Principal mandate:

- operate as a centre of research, standardisation and dissemination of techniques for controlling animal diseases;

- to place expert consultants at the disposal of the OIE.
OIE Collaborating Centres

May also contribute to:
- provide technical training to personnel from OIE Members;
- organise scientific meetings on behalf of the OIE;
- coordinate scientific and technical studies;
- publish any information in their competence that may be useful to OIE Members.
OIE Collaborating Centres for Aquatic Animal Diseases

OIE Collaborating Centre for Information on Aquatic Animal Diseases
The Centre for Environment, Fisheries & Aquaculture Science (CEFAS), UK

OIE Collaborating Centre for Epidemiology and Risk Assessment of Aquatic Animal Diseases
Canada and Norway – adopted at GS in May 2010

Contact details of RL’s and CC’s are listed in the Aquatic Manual
The main objective

To assist laboratories in developing or in-transition countries to build their capacity and scientific expertise

Easier access to expertise for the rapid detection and diagnosis of disease

To achieve an even geographical distribution of expertise and Reference Laboratories.
Each Twinning Project..

Is a link between OIE Reference laboratory or Collaborating Centre (Parent) and national laboratory (Candidate).

Aims to improve expertise and diagnostic capacity with eventual aim of providing support within the region and reaching OIE standards.
Laboratory Twinning Projects for Aquatic Animal Diseases

Several twinning project proposals for aquatic animal diseases are under consideration by the OIE (will be the first);

OIE members encouraged to consider opportunities for future laboratory twinning projects
Twinning Guide and Project Template/Contract

- Background
- How to apply/proposal
- Basic project management
- Project plan (objectives, stages)
- Regular monitoring
- Facilitates efficient applications
- Formalises agreement

http://www.oie.int/downld/LABREF/A_Guide.pdf