



The EU Diagnostic Manuals

www.eurl-fish.eu -

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A new Commission Decision is in preparation

Draft
COMMISSION DECISION
of
implementing Directive 2006/88/EC as regards requirements for surveillance and diagnostic
methods
(Text with EEA relevance)

THE EUROPEAN COMMISSION,
Having regard to the Treaty on the Functioning of the European Union,
Having regard to Council Directive 2006/88/EC of 24 October 2006 on animal health
requirements for aquaculture animals and products thereof, and on the prevention and control
of certain diseases in aquatic animals¹, and in particular Article 40(1), 50(4) and 61(3) thereof;
Whereas
(1) Directive 2006/88/EC provides for certain preventive measures relating to the
surveillance and early detection of the aquatic animal diseases listed in Annex IV to
that Directive and certain measures to be applied in the event of suspicion of, or an
outbreak of those diseases in aquatic animals;
(2) Directive 2006/88/EC provides for certain requirements to achieve the status of
disease free Member States, zones or compartments;
(3) It is necessary to lay down at European Union level diagnostic procedures, sampling
methods and criteria for the evaluation of the results of laboratory tests that may lead
to a suspicion, confirmation or to achieve the status of disease free zone or
compartment;
(4) A sufficient period of time should be provided for the implementation of these
measures;
(5) All measures described in this Decision should harmonise with the Manual of
Diagnostic Tests for Aquatic Animals of the World Animal Health Organisation (OIE);
(6) The measures laid down in this Decision are in accordance with the opinion of the
Standing Committee on the Food Chain and Animal Health;

HAS ADOPTED THIS DECISION:

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ANNEX PART I

Introduction and objectives

1. In order to ensure uniform procedures for surveillance schemes, including health inspections and sampling, and diagnosis of the aquatic diseases listed in Annex IV to Directive 2006/88/EC, this Decision sets out:
 - (a) minimum requirements for surveillance schemes and diagnostic methods that shall be used by Member States to achieve, regain or maintain disease free status as regards the aquatic diseases listed in Annex IV to Directive 2006/88/EC for the Member State, zones or compartments;
 - (b) minimum requirements and criteria for the evaluation of the results for diagnostic methods to be performed in the case of suspicion and to confirm the presence of the listed diseases;
2. This Decision is directed towards both the authorities responsible for the control of those diseases and the laboratory personnel performing the tests with regard to those diseases.
3. The confirmation of the listed diseases in aquatic animals must be in accordance with the criteria for the evaluation of the results for diagnostic methods set out in this Decision.
4. For the purpose of this Decision, in addition to the definitions laid down in Directive 2006/88/EC, the following definitions shall apply:
 - (a) 'category I' means disease free in accordance with Articles 49 or 50 of Directive 2006/88/EC;
 - (b) 'category II' means subject to a surveillance programme established in accordance with Article 44(1) of Directive 2006/88/EC;
 - (c) 'category III' means an undetermined health status, i.e. neither covered by categories I, II, IV nor V;
 - (d) 'category IV' means subject to an eradication programme established in accordance with Article 44(2) of Directive 2006/88/EC;
 - (e) 'category V' means subject to containment measures according to Article 38 of Directive 2006/88/EC.

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PART II: SURVEILLANCE AND DIAGNOSTIC METHODS FOR VHS AND IHN

I. Aetiology of VHS and IHN

II. Provisions for programmes to achieve and to maintain certain health statuses with regard to VHS and/or IHN and to contain VHSV and/or IHNV infections

- II.1. General requirements for inspections and sampling
- II.2. Specific provisions to achieve disease free health status (category I) with regard to VHS and/or IHN
 - II.2.1 Surveillance programmes:
 - II.2.2 Eradication programmes
- II.3. Specific provisions for maintenance of category I status with regard to VHS and/or IHN

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PART III: SURVEILLANCE AND DIAGNOSTIC METHODS FOR EHN

5

As exotic disease diagnostic methods only. Follows the same as for VHS/IHN

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PART III: SURVEILLANCE AND DIAGNOSTIC METHODS FOR KHVD

6

Follows the same as for VHS/IHN but only in freshwater

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Samples shall be tested in accordance with the approved methods as described on the EURL website.

VHS/IHN as example:



1 Decision comprising all listed fish and molluscs diseases.
Detailed diagnostic procedures on web sites and close to OIE guidelines

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• **ANNEX II ,PART I:**
_SURVEILLANCE AND DIAGNOSTIC METHODS FOR VHS AND IHN

- I. Aetiology of the diseases in question
- II. Programmes to achieve and to maintain category 1 status
- III. Diagnostic methods

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II.2. Specific provisions to achieve disease free health status (category I) with regard to VHS and/or IHN

II.2.1 Surveillance programs:

(a)(i) Model A – two-year surveillance program:

LARGE sample size:
2 year x 2 x 150 fish

In all 600 fish

TABLE II A
Surveillance scheme for zones and for compartments for the two year control period which precedes achievement of VHS and/or IHN free status

	Number of health inspections per year (two years)	Number of samplings per year (two years)	Number of fish in the sample ¹	
			Number of growing fish	Number of broodstock fish
(a) Farms with broodstock	2	2	120 (first) 150 (second)	0 (first inspection) 0 (second inspection)
(b) Farms with broodstock only	2	1	0	150 (first or second inspection)
(c) Farms without broodstock	2	2	150 (first and second inspection)	0

Maximum number of fish per pool: 10
¹ The samples have to be collected no sooner than three weeks after transfer of fish from fresh to saltwater.

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II.2. Specific provisions to achieve disease free health status (category I) with regard to VHS and/or IHN (cont)

(ii) Model B – 4 years surveillance programme with reduced sample size:

SMALL sample size:
2 year x 1 x 30 fish +
2 year x 2 x 30 fish
In all 180 fish samples

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TABLE II B
Surveillance scheme with reduced sample size for the four year control period which precedes achievement of VHS and/or IHN free status

	Number of health inspections per year	Number of samplings per year	Number of fish in the sample ¹	
			Number of growing fish	Number of broodstock fish ²
First two years of the surveillance period				
(a) Farms with broodstock	2	1	0 (first inspection) 30 (second inspection)	0 (first inspection) 0 (second inspection)
(b) Farms with broodstock only	2	1	0	30 (first or second inspection)
(c) Farms without broodstock	2	1	30 (first or second inspection)	0
Last two years of the surveillance period				
(a) Farms with broodstock	2	2	30 (first inspection) 0 (second inspection)	0 (first inspection) 30 (second inspection)
(b) Farms with broodstock only	2	2		30 (first and second inspection)
(c) Farms without broodstock	2	2	30 (first and second inspection)	

Maximum number of fish per pool: 10
¹ The samples have to be collected no sooner than three weeks after transfer of fish from fresh to saltwater.

II.2.2 Eradication programmes

- II.2.2.1 General requirements

Protection zone

- in coastal areas: at least 5 km,
- in inland areas: the entire water catchment area; but possibility for derogation

Surveillance zone

- in coastal areas: radius 10 km
- in inland areas: as an extended area outside the established protection zone.

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II.2.2 Eradication programmes (cont.)

The following period shall be at least 6 weeks. When all farms officially declared infected are emptied. Synchronised following shall be carried out (for ISA longer period up to 6 months)

Restock with fish from category I only

Surveillance schemes before approval



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II.3.
Specific provisions for maintenance of category I status

Maintenance programs:

Sample size
1 x 30 fish

TABLE II C
 Surveillance schemes for zones or compartments to maintain VHS and/or IHN free status

Risk level	Number of health inspections	Number of fish in the sample
High	1 every year	30 ¹
Medium	1 every 2 years	30 ²
Low	1 every 4 years	30 ³

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III. Diagnostic methods
 VHS, IHN, and EHN



III.1. Organs to be sampled and Diagnostic methods for VHS, IHN, and EHN:

- Spleen, anterior kidney, and either heart or encephalon.
- Maximum 10 fish may be pooled.
- virus isolation on cell culture followed by identification using virus neutralisation test, IFAT, ELISA or RT-PCR.
- Real-time RT-PCR for VHS and IHN included in 2012.

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III. Diagnostic methods

ISA



III.1. Organs to be sampled and Diagnostic methods for ISA:

- Diagnosis is based on a combination of pathological findings, cell cultivation and immunochemical and or molecular testing
- RT-PCR: Anterior or mid-kidney + heart
- Cell cultivation: Liver, anterior kidney, heart and spleen
- Histology: Liver, spleen, mid-kidney, heart, pancreas and gills
- Maximum 5 fish may be pooled.
- Virus isolation on cell culture (ASK-1, TO) followed by identification using IFAT or RT-PCR.

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III. Diagnostic methods

KHV



III.1. Organs to be sampled and Diagnostic methods for KHV:

- Diagnosis is based on finding of CyHV-3 by PCR and eventually by cell cultivation.
- PCR: gill, kidney and spleen tissue
- Cell cultivation: gill, kidney and spleen tissue
- No pooling or max. 5 fish may be pooled when clinical symptoms.
- Virus isolation on CCB cell culture followed by identification using IFAT or PCR.

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III. Diagnostic methods
EUS

III.1. Organs to be sampled and Diagnostic methods for *Aphanomyces invadans*:

Sampling should be done from various lesions and organs and should be done in the edge of a lesion from a clean cut surface



EUS-affected grey mullet *Mugil cephalus*, caught in the Richmond River, eastern Australia.

Conclusions

Final decision should be settled within too long
Still remaining legal questions on keeping legislative topics on websites outside the Commission

- No much changes from previous legislation
[Commission Decision 2001/183/EC](#) (VHS/IHN) and
[Commission Decision 2003/466/EC](#) (ISA)

No previous for KHV and EHN

Real-time RT-PCR included for surveillance and diagnostics of VHS, IHN and ISA