



ENZYME IMMUNOASSAYS FOR THE DIAGNOSIS OF FISH INFECTIOUS DISEASES

Infectious Pancreatic Necrosis Virus (IPNV)
Spring Viraemia of Carp Virus (SVCV)
Viral Hemorrhagic Septicaemia Virus (VHSV)

ELISA kits are optimized and validated for detection of the viral antigen
in fish organ homogenates and in culture media of infected cell cultures

Introduction



Infectious pancreatic necrosis virus (IPNV)

IPNV is a birnavirus, many types of which infect marine finfish and shellfish. The signs below are of the virulent disease in salmonids.

Clinical signs: fish lie still on bottom, fish swim with a spiralling motion, white faecal casts, swollen belly, darkening body colour, exophthalmus (pop eye), lesions and ulcers in pancreas, oesophagus and stomach, intestines empty or filled with clear mucus.

Spring viraemia of carp virus (SVCV)

SVCV is a rhabdovirus closely related to the infectious haematopoietic necrosis virus and the haemorrhagic septicaemia virus. It is the cause of contagious infection seen in many species of cyprinids around 1 or 2 years of age, but other freshwater fishes are also susceptible. The disease is widespread in European countries but has more recently appeared in Asia and the Americas.

Clinical signs: Exophthalmus (pop eye), swollen abdomen (dropsy), petechial (pinpoint) haemorrhages in the fatty tissue and muscle surrounding organs and stomach wall, haemorrhages on skin, haemorrhages in gills, abdominal tissue, swim bladder and other internal organs, ascites (abdominal cavity filled with fluid).

Viral hemorrhagic septicaemia virus (VHSV)

VHSV is a rhabdovirus. Several genogroups/genotypes of the virus have been identified from different environments in different parts of the world.

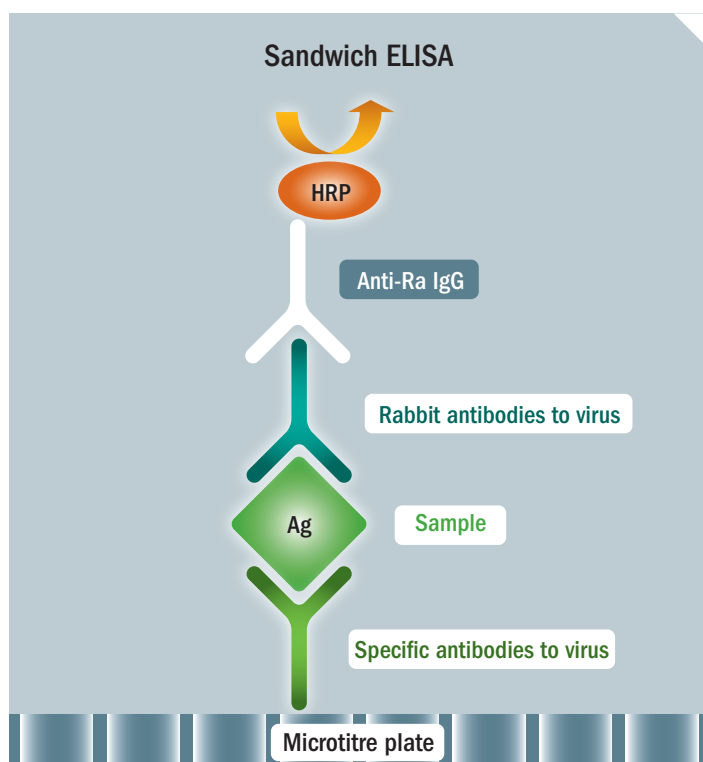
Clinical signs: Some fish show no external symptoms, but others show signs of infection that include bulging eyes, bloated abdomens, bruised-looking reddish tints to the eyes, skin, gills and fins. Internally, the virus can cause petechial haemorrhaging (tiny spots of blood) in internal muscle tissue, and petechial or severe haemorrhaging in internal organs and other tissues (liver, kidneys, spleen, and skeletal muscle). There may also be a nervous form of the disease where fish are constantly flashing and showing abnormal behaviour.

Clinical Application

- ▶ Detection of viruses in organ homogenates and in the media of infected cell cultures
- ▶ Diagnosis of the infection
- ▶ Control of fish before transport
- ▶ Control of quarantined fish (export, import)

Test Principle











The assays are based on a sandwich type of ELISA method.



User Comfort

- ▶ Ready-to-use components
- ▶ Colour-coded components
- ▶ Interchangeable components
- ▶ Breakable colour-coded microplate strips
- ▶ Negative antigen, Positive antigen and Rabbit serum containing antibodies to virus included
- ▶ Easy assay procedure

Summary of EIA Protocol (Rapid test)

Step No.	Test steps
1	 Dilute samples organ homogenates, culture media of infected cell cultures
2	 Pipette 50 µl Rabbit serum containing antibodies to virus and 50 µl Control Antigens and diluted samples
3	 Incubate 60 minutes at 37°C
4	 Aspirate and wash wells 4 times
5	 Add 100 µl Conjugate
6	 Incubate 60 minutes at 37°C
7	 Aspirate and wash wells 4 times
8	 Add 100 µl Substrate (TMB-Complete)
9	 Incubate 10 minutes at room temperature
10	 Add 100 µl Stopping solution
11	 Read colour intensity at 450 nm

Total assay time according to duration of sample incubation:
a) overnight incubation: second day results
b) rapid test: approximately 2.5 h

Test Characteristics

ELISA	Assay Sensitivity
IPNV Ag ELISA	10 ² TCID ₅₀ per 0.1 ml of the fluid examined
VHSV Ag ELISA	10 ³ TCID ₅₀ per 0.1 ml of the fluid examined
SVCV Ag ELISA	10 ^{2,8-3,5} TCID ₅₀ per 0.1 ml of the fluid examined

Advantages

- ▶ Identical assay procedure
- ▶ High diagnostic specificity and sensitivity
- ▶ High reproducibility
- ▶ Short total assay time (possible)
- ▶ Ready for automation
- ▶ Customer support

VETERINARY DIAGNOSTICS

Ordering Information

Cat. No.	Product	No. of Tests
IPN096	IPNV Ag ELISA	96
SVC096	SVCV Ag ELISA	96
VHS096	VHSV Ag ELISA	96

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Company is certified to the quality management system standards ISO 9001 and ISO 13485 for in vitro diagnostics.