



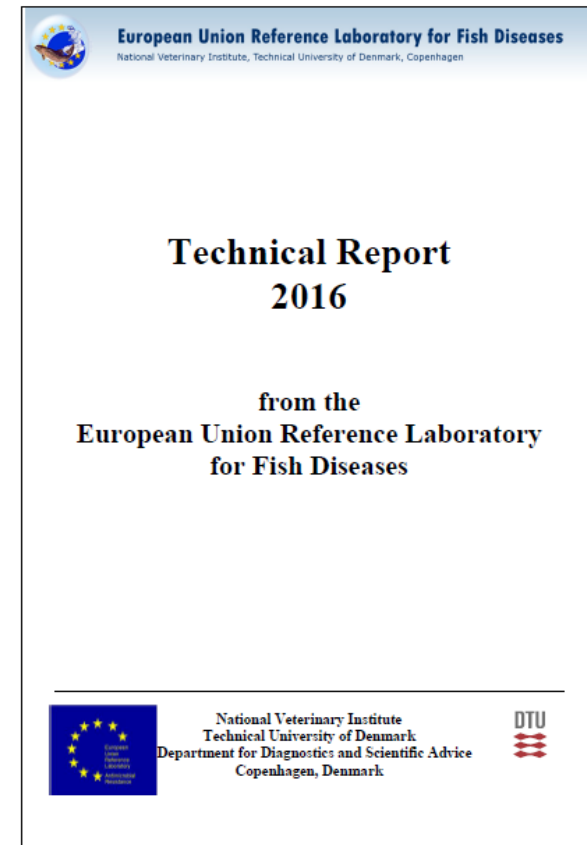
European Union Reference Laboratory for Fish Diseases

National Veterinary Institute, Technical University of Denmark, Copenhagen



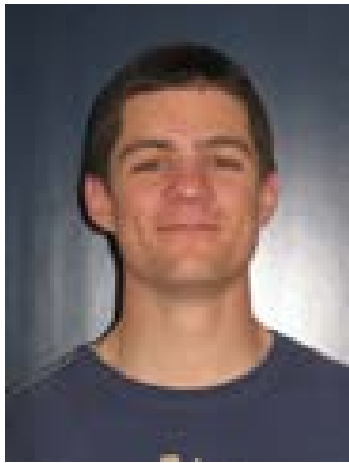
EURL-Fish work done in 2016

Niels Jørgen Olesen





Niels Jørgen
Olesen



Niccoló
Vendramin



Argelia Cuenca
Navarro



Tine Iburg



Lone Madsen



Jacob G., Schmidt



Nikolaj G. Andersen



Anna L. F. Alencar



National Veterinary Institute
Didde
Sørensen



Betina
Lynnerup



Christina Flink
Desler



Troels Secher
Rundqvist

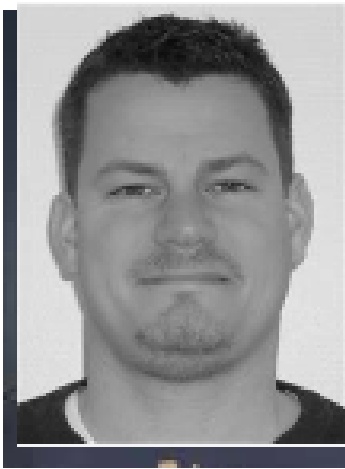


Teena Klinge





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Teena Klinge



EURL-Fish work program 2016

5 main objectives

- **1. Coordination and training**
- **2. Proficiency test**
- **3. Reagents and products**
- **4. Scientific advice and activities**
- **5. Missions**

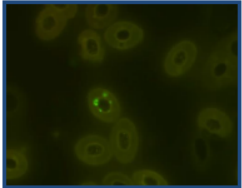
20 subgoals

1-1,1-2 Organise and prepare for the 20th Annual Meeting for the NRLs and produce a report from the Annual Meeting

 **European Union Reference Laboratory for Fish Diseases**
National Veterinary Institute, Technical University of Denmark, Copenhagen

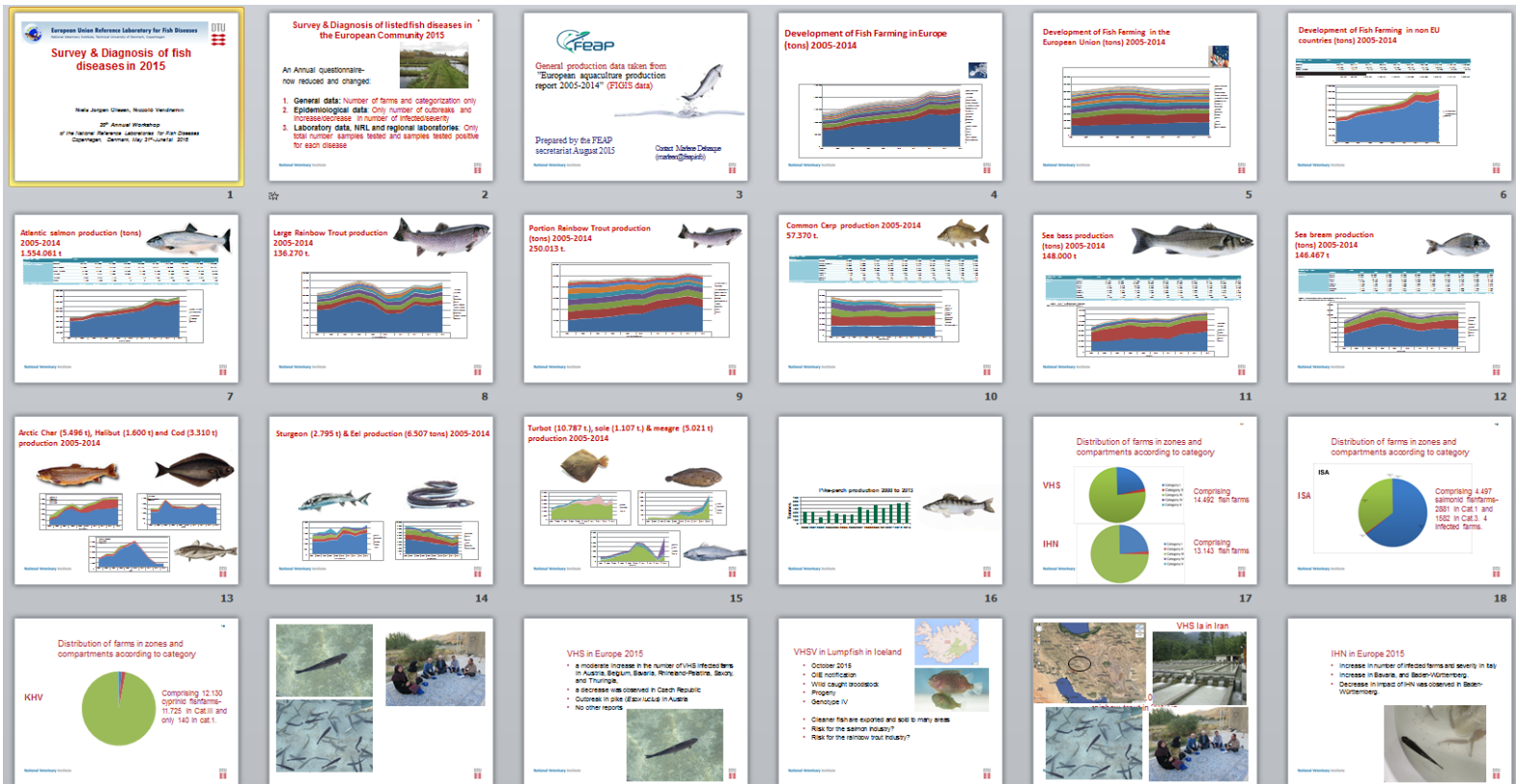
Report:
**20th Annual Workshop of the
National Reference Laboratories
for Fish Diseases**

**Copenhagen, Denmark
May 31st – June 1st 2016**

		
Red mark syndrome in Rainbow trout	Atlantic Salmon Red blood cells with intracytoplasmic inclusions	Lumpfish (<i>Cyclopterus lumpus</i>)

Organised by the European Union Reference Laboratory for Fish Diseases
National Veterinary Institute, Technical University of Denmark

1-3 Collect and report data on the fish diseases situation in EU



1-4 Facilitate and provide training in laboratory diagnosis:

EURL training courses 2016

EURL course 1: “Methods for implementation of surveillance procedures for listed fish diseases” took place from 5th to 9th October 2016

EURL course 2: “Introduction to histopathology in fish diseases” and took place in Copenhagen 12th to 15th October 2015

Training courses:



2-1, 2-2 Prepare and report the Annual Inter-laboratory Proficiency Test.

PT1-2014 for identification of: VHSV, IHNV, EHNV, SVCV and IPNV

and PT2 identification of: CyHV-3 (KHV), SAV and ISAV



EURL for Fish Diseases

Report of the Inter-Laboratory Proficiency Test 2016

for identification and titration of

VHSV, IHNV, EHNV, SVCV and IPNV (PT1)

and identification of

CyHV-3 (KHV), SAV and ISAV (PT2)

Organised by the
European Union Reference Laboratory for Fish Diseases,
National Veterinary Institute, Technical University of Denmark,
Copenhagen, Denmark

3-1 Supply reference reagents to the NRLs in Member States

Materials supplied by the EURL

On request, the EURL supplies material to other laboratories in Member States and Third Countries to aid in the diagnosis and characterisation of fish diseases.

Annex 1 Reagents supplied by the EURL-Fish in 2016

Country	Name	Institute	Date of receipt	Material	Amount	No.
The Netherlands	Olga L.M. Haenen	Central Veterinary Institute (CVI) of Wageningen UR	20.01.16 + 26.01.16	Freeze dried VHSV	1 ampoule	
Italy	Anna Toffan	Istituto Zooprofilattico Sperimentale delle Venezie	20.01.2016	Cell supernatant. Virus not identified	2 tubes	15-16934
Norway	Bjorn Spillberg	Norwegian Veterinary Institute	26.01.2016	Blood infected with Virus-Y	2 tubes	14-4566
Iceland	Sigrður Gudmundsdóttir	Institute for Experimental Pathology University of Iceland	08.02.2016	Antibodies: IPN, VHS, IHN, SVC	6 tubes	
Spain	Pilar Fernández Somoza	Laboratorio Central de Veterinario MAGRAMA	11.02.2016	Antibodies, VHS + IHN	2 tubes	
The Netherlands	Olga L.M. Haenen	Central Veterinary Institute (CVI) of Wageningen UR	07.03.2016	EPC and FHM cells	2x 2 flasks	
The Netherlands	Olga L.M. Haenen	Central Veterinary Institute (CVI) of Wageningen UR	29.03.2016	BF-2 cells	4 flasks	
Canada	Dante Mátwo	Atlantic Veterinary College University of Prince Edward Island (UPEI)	07.04.2016	SAV II SAV VI	2x 2 tubes	9895349 9895379
Scotland	Eann Munro	Diagnostic Group Leader Marine Scotland – Science Scottish Government	12.04.2016	G24-IHNV panel SHRV HIRRV	24 tubes 1 tube 1 tube	16-6904 16-5313
Korea	Hyoung Jun KIM	NFQS National Fishery Products Quality Management Service	15.04.2016	BF-2 cells	1 flask	
Italy	Anna Toffan	Istituto Zooprofilattico Sperimentale delle Venezie	20.04.2016	Supernatant of organs homogenate from Sturgeon	2x 1 tube	16-5934
Brazil	Anna Luiza Farias Alencar	Universidade de São Paulo FZEA	20.04.2016	CHSE cells	2 small flasks	
Estonia	Ave-Ly Toomvapp	Estonia Veterinary and Food Laboratory	24.05.2016	BF-2 and EPC cells	2x 2 small flasks	
The Netherlands	Olga L.M. Haenen	Central Veterinary Institute (CVI) of Wageningen UR	01.06.2016	BF-2 cells	2 flasks	
Poland	Marek Matras	National Veterinary Research Institute	01.06.2016	CHSE cells	2 flasks	

Country	Name	Institute	Date of receipt	Material	Amount	No.
Belgium	François Liefthj	CEER/Groupes Fish Diseases Laboratory	06.06.2016	EPC-1 cells	2 flasks	
Belgium	A. Vandaplasschen	University of Liège	06.06.2016	Virus from Eel	5 tubes	
Sweden	Charlotte Assin	National Veterinary Institute	06.06.2016	Supernatant after grinding of organs from Herring	9 tubes	16-4756
The Netherlands	Olga L.M. Haenen	Central Veterinary Institute (CVI) of Wageningen UR	08.06.2016	EPC cells	2 flasks	
UK - England	Richard Keith Poley	CEFAW Weymouth Laboratory	08.06.2016	KF-1 cells	2 flasks	
Italy	Anna Toffan	Istituto Zooprofilattico Sperimentale delle Venezie	22.06.2016	Cell supernatant. Virus not identified	2 tubes	16-7445
Norway	Torfinn Meldal	Norwegian Veterinary Institute	28.06.2016	G24-IHNV panel Infected cell supernatant	24 tubes + 1 tubes	15-16994
Italy	Anna Toffan	Sperimentale delle Venezie	06.07.2016	Fish tissue	6+1 tubes	16-10588
Chile	Ricardo Enriquez Soto	Facultad de Ciencias Veterinarias Universidad Austral de Chile	19.07.2016	FTA Cards (VHSV) EPC cells BF-2 cells	2x 4 cards 2 flasks 2 flasks	
Italy	Anna Toffan	Istituto Zooprofilattico Sperimentale delle Venezie	24.08.2016	Cell supernatant. Virus not identified. RTG-2 cells KF-1 cells	4 tubes 1 flask 2 flasks	16-7450 16-9507
India	Kooloth Valappil Rajendran	Indian Council of Agricultural Research (ICAR)	24.08.2016	FTA Cards (VHSV)	2 cards	
Norway	Espen Ramstad	Veterinærhøyskolen - NMBU	14.09.2016	PRV-On infected blood	9 tubes	15-4566
Serbia	Vladimir Ivan Radovic/jevic	Institute of Veterinary Medicine of Serbia	14.09.2016	BF-2 cells EPC cells CHSE cells	2 flasks 2 flasks 2 flasks	
Norway	Maria Dabla	Norwegian Veterinary Institute	03.10.2016	rNA	118 tubes	15-10833
UK - England	Keith Way	CEFA Weymouth Lab	03.10.2016	Mab anti IHNV	1 tube	
Belgium	François Liefthj	CEER/Groupes Fish Diseases Laboratory	03.10.2016	SAV VI	1 ampoule	
France	Yannick Blanchard	ANSES Plouzanet Pisciculture Laboratory	12.10.2016	VHSV infected cell supernatant	13 tubes	16-10894
Norway	Maria Dabla I	Norwegian Veterinary Institute	17.10.2016	Full blood (PRVom)	15 tubes	16-15041

Country	Name	Institute	Date of receipt	Material	Amount	No.
Korea	Hyun-Wook KWON	NFQS National Fishery Products Quality Management Service	26.10.2016	VHSV infected supernatant VHSV Ring Test PCR Product	25 tubes 2x 5 FTA Cards 24 tubes	
P.R. China	Liu Hong	The National Key Laboratory of Aquatic Animal Health Animal and Plant Inspection and Quarantine Centre, Shenzhen Exit/Entry Inspection and Quarantine Bureau General AQSIQ	04.11.2016	EPC cells BF-2 cells CHSE cells SSN cells	2 flasks 2 flasks 2 flasks 2 flasks	
Norway	Karune Lindmo	PHARMAQ AS	07.11.2016	IHNV, cell culture	3 tubes	
Denmark	Torsten Boutrup	Aquapri A/S	14.11.2016	Eggs; MEM Glass slides (Atlantic salmon with PRV and IHNV)	1 flask 136 slides	
Norway	Anne Bent Olsen	Norwegian Veterinary Institute	16.11.2016	CCB cells SSN-1 cells GF cells WSSK cells	2 flasks 2 flasks 2 flasks 2 flasks	
Norway	Simon Wei	Norwegian Veterinary Institute	16.11.2016	CCB cells SSN-1 cells GF cells WSSK cells	2 flasks 2 flasks 2 flasks 2 flasks	
China	Tao Sun	Shandong Entry-exit Inspection and Quarantine Bureau	22.11.2016	Mab IP5B11 MAb IHNV Hyb 136-3	1 tube 1 tube	
Serbia	Vladimir Ivan Radovic/jevic	Institute of Veterinary Medicine of Serbia	30.11.2016	CCB cells CHSE cells	2 flasks 2 flasks	
Norway	Maria Dabla	Norwegian Veterinary Institute	30.11.2016	Fish tissue (PRV-IHNV)	118 tubes	15-10833
Peru	Mervin Guevara Torres	Coastal Laboratory of IMARPE, Tumbes headquarters	07.12.2016	FTA Cards - VHSV	2 cards	
Denmark	Time Hbug Heike Schütze	NVI - DTU FLI	07.12.2016	VHSV PT2016 on FTA Cards	5 cards 5 cards 5 cards 5 cards 5 cards	
France	Thierry Morin	ANSES	07.12.2016	VHSV PT2016 on FTA Cards	5 cards 5 cards 5 cards 5 cards	
Italy	Anna Toffan	IZSVe	07.12.2016	VHSV PT2016 on FTA Cards	5 cards 5 cards 5 cards	
England	David Stone	CEFAW S	07.12.2016	VHSV PT2016 on FTA Cards	5 cards 5 cards 5 cards	
Korea	Kwon Hyun	NFQS	07.12.2016	VHSV PT2016 on FTA Cards	5 cards 5 cards 5 cards	
Norway	Knut Falk	Norwegian Veterinary Institute	19.12.2016	Mab IHNV	1 tube	

3-2 Production of antisera against selected isolates when necessary

In 2016 no new productions of antisera were needed and our stocks of supernatants from hybridoma cells producing monoclonal antibodies were sufficient for the year

3-3 Update and maintain a library of isolates of ISAV, VHSV and IHNV, KHV and EHN

- Several isolates of the listed viruses VHSV, IHNV and KHV were received and stored in our library during 2016.

Annex 2 Reagents received in 2016

Country	Name	Institute	Date of receipt	Material	Amount	Protocol No.
Belgium	François Lieféig	CER Groupe Fish Diseases Laboratory	26.01.16	Sperm + Coelomic liquid	1 + 2 tubes	16-2045
Sweden	Astrid Fishexport AB		22.03.2016	Whole herrings	175 fish	16-4758
Italy	Anna Toffan	Istituto Zooprofilattico Sperimentale delle Venezie	19.04.16	WSSK cells	1 small flask	
Croatia	Snjezana Zrncic	Veterinary Institute Lab. of Fish and Molluscs Pathology	28.04.16	Organs Seabass	13 samples	16-7030
Slovenia	Vlasta Jencic	University of Ljubljana, Veterinary Faculty	18.05.16	Kidney	30 units	16-7744
Italy	Anna Toffan	Istituto Zooprofilattico Sperimentale delle Venezie	27.05.16	Freeze dried inactivated viral supernatant	6 vials	16-8497
Croatia	Snjezana Zrncic	Veterinary Institute Lab. of Fish and Molluscs Pathology	17.06.16	Homogenate organ supernatant	6 samples	16-9625
Serbia	Vladimir, Ivan Radosavljevic	Institute of Veterinary Medicine of Serbia	02.08.16	Homogenate carp tissue (gills and Kidney). Homogenate trout tissue.	3 samples 2 samples	16-12103
Japan	Takafumi Ito		08.09.16	Monoclonal HIRRV antibodies	4 tubes	16-14572
Norway	Torfinn Moldal	Norwegian Veterinary Institute	14.09.16	Pox-control	2 tubes	16-14800
Norway	Ole Bendik Dale	Norwegian Veterinary Institute	27.09.17	Pox-antibodies Pox pos. histoslides Pox pos. Paraffin blocks	1 tube 4 slides 2 blocks	16-15593
Austria	Mansour El-Matbouli	Veterinärmedizinische Universität Wien	26.10.16	Homogenated organs + DNA	3 samples 3 samples	16-17468
Scotland	Hannah Stagg	Disease Diagnostic Dept. Marine Scotland Science Marine Laboratory	08.11.16	Cell supernatant	2 tubes	16-18335

Country	Name	Institute	Date of receipt	Material	Amount	Protocol No.
Ireland	Neil Martin Ruane	Marine Institute Fish Health Unit Rinville, Oranmore	23.11.16	Cell supernatant	2 tubes	16-19667
Iceland	Sigrídur Guðmundsdóttir	Institute for Experimental Pathology University of Iceland	13.12.16	Cell supernatant	3 vials	16-20738

4-1 Update the webpage for the EURL, www.eurl-fish.eu

The EURL website (www.eurl-fish.eu) is a notice board, where NRLs and other interested parties can access relevant information and previous reports concerning the activities coordinated by the EURL and relevant upcoming events in the Union.

European Union Reference Laboratory for Fish Diseases
National Veterinary Institute

ACTIVITIES REPORTS MANUALS NRL NETWORK LEGISLATION LINKS NEW

What is the EURL?
The European Union Reference Laboratory (EURL) by the European Commission and is situated at Institute in Denmark. The functions and duties ; diagnostic procedures for notifiable fish disease

The functions and duties of the European Commission Fish Diseases are described in [Council Directive](#) of the EURL is to ensure the quality of diagnostic States and to harmonise the procedures and m is mainly concerned with the exotic and non-ex in [Council Directive 2006/88/EC](#).

The EURL co-ordinates those activities of the h (NRLs) for Fish Diseases in EU that aim to harm and disseminate information of mutual interest, is decided at the Annual Meeting of the NRLs fi

National reference laboratories 2 / 4

NEWS All **CALENDAR** All **The Dat**

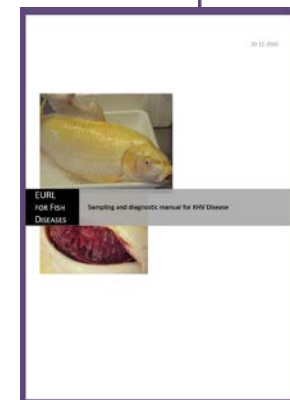
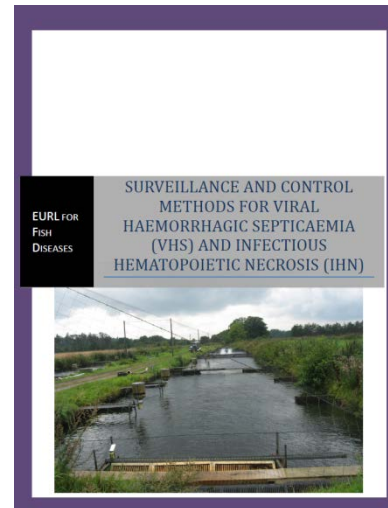
24 March 2017
Proficiency Test 2016

24 March 2017
21st Annual Workshop

20 February 2017
Questionnaire on Survey and Diagnosis of Fish Diseases in Europe 2016

4-2 Update the diagnostic manuals for VHS, IHN, ISA, KHV disease, and EHN on the EURL web page

- Commission Decision 2015-1554 finally adopted and implemented
- Easy access through our web page



4-3 Fishreflabnet: Maintain and further develop the interactive network with the NRLs, Fishreflabnet , in order to promote a more proactive data sharing and communication with and between reference laboratories in member states.

- This tool is used for newsletters, scientific updates and announcements from the EURL Fish like announcements and invitations for the Annual Workshop or publication of content in the ampoules from the proficiency test or on the final Inter-laboratory Proficiency test report. In addition the e-mail group is used for announcing other workshops, training courses and conferences and new publications of interest for the NRL Fish network

4-4 Identify and characterise selected isolates of listed viruses

- In 2015 a number of virus isolates, sera and other reagents were received for further characterisation at the EURL
- and for storing in our virus library

Member States and countries outside EU		
Material received	Laboratories	Units
Diagnostic material for virology	8	210 samples
Diagnostic material for PCR	8	45 samples
Diagnostic material for bacteriology	1	30 samples
Cell cultures	1	1 sample
MAB/PAb	1	4 samples
Other material	1	7 samples

4-5 Update and expand www.fishpathogens.eu with more pathogens

The database now consisted in 1194 VHSV records, of those 811 are public, and the rest are placed as restricted. Both betanoda and IHNV databases have numbers similar to last year, with 96 records for IHNV and 62 for betanoda. A new SAV database has been established. A number of new features are in the process to be added to all databases. A number of bugs were also corrected.

The screenshot shows the homepage of the Fish Pathogens Database. At the top is a blue navigation bar with links for Home, About, Contact, Terms and Conditions, F.A.Q., How to cite, Create an account, and Log in. Below the navigation bar is the site logo, which consists of a stylized DNA double helix and the text 'FISH PATHOGENS'. The main heading is 'Fish Pathogens Database'. Below this, there is a paragraph explaining that the platform is for sharing information on fish pathogen isolates and sequences. It mentions that the databases are free to use but require a subscription. There are also links for 'Terms and Conditions' and 'F.A.Q.'. At the bottom of the screenshot, there are three blue buttons labeled 'VHSV >', 'IHNV >', and 'Betanodavirus >'. Below these buttons are three small images: a fish, a circular diagram, and a close-up of a fish's head.



4-6 Perform molecular epidemiology analysis to improve knowledge on diseases spreading mechanisms of viral pathogens

- A standardization and validation of the **IHNV** real-time RT-PCR developed by Purcell et al. was initiated in 2016. The method is being translated and validated to a one-step procedure which is more convenient for use as a tool for surveillance of IHNV.
- A real-time PCR for detection of **Salmon pox-virus** and for Cyprinid edema virus (CEV) was implemented.
- In addition we have implemented a two-step RT-qPCR to detect **Atlantic salmon Calicivirus** and a conventional PCR for detecting ***Onchorynchus mykiss virus (OMV)***.
- In addition, we implemented and are currently testing a real-time PCR for detecting ***Renibacterium salmoninarum*** the causative agent of bacterial kidney disease (BKD). We have also implemented a qPCR to detect and quantify ***Tetracapsuloides bryosalmonae*** the causative agents of proliferative kidney disease (PKD).

4-8 Emerging diseases:

Red Mark Syndrome (RMS) As presented earlier by Jacob G. Schmidt
Salmon gill poxvirus (SGPV)
Piscine orthoreovirus (PRV)

4-10 Molecular characterization of fish cell lines: Perform molecular analysis to “barcode” and certify cell lines routinely used for viral diagnostics

Cell line	Name	Expected origin	De Facto origin
EPC*	<i>Epithelioma Papullosum Carpio</i>	Common carp	Fat Head Minnow
BF-2	Bluegill Fry	Bluegill	Green sunfish / bluegill
CHSE-214	Chinook Embryo	Salmon	Chinook Salmon
RTG-2	Rainbow trout gonad	Rainbow trout	Rainbow trout
FHM	Fat Head Minnow	Fat Head Minnow	Fat Head Minnow
CCO	Channel Catfish Ovary	Channel Catfish	Brown bullhead
EK-1	Eel Kidney	Pacific eel.	Japanese eel
ASK	Atlantic Kidney	Salmon	Atlantic Salmon
CCB	Common Carp Brain	Common Carp	Common Carp
SBL	Sea Bass Lymphoid	European seabass	Chinook Salmon
WSSK	White Sturgeon Skin-1	White sturgeon	white sturgeon

5-1 Missions: Organizing missions to relevant laboratories. Missions will focus on NRLs where on-site communication would be beneficial. As collaboration with NRLs in 3rd countries from where EU is importing large amount of fish

Visit in Korea, and China

Visit to Switzerland

5-2 Attending missions, international meetings and conferences in order to be updated on emerging and listed fish diseases.

22nd Annual Workshop 2018

- When?
- Where?

