

### First detection of Piscine Orthoreovirus genotype 3 (PRV-3) in the Netherlands at a rainbow trout farm, July 2022

Olga Haenen, Tine Moesgaard Iburg, Michal Voorbergen-Laarman, Betty van Gelderen, Marc Engelsma, Argelia Cuenca, Niccolò Vendramin, Niels Jørgen Olesen

WBVR, NRL for Fish Diseases of the Netherlands, with help of EURL for Fish Diseases



1

Casus in July 2022: Rainbow trout in pond of culture farm in Central NL

- 40-50% mortality in pond rainbow trout of 15-27 cm, in short time
- Water temperature 18°C
- Less flow-through because of algae during weeks
- Fish originated in January 2022 from a farm in the very South of NL (imported eyed eggs from Denmark with a health certificate)



2

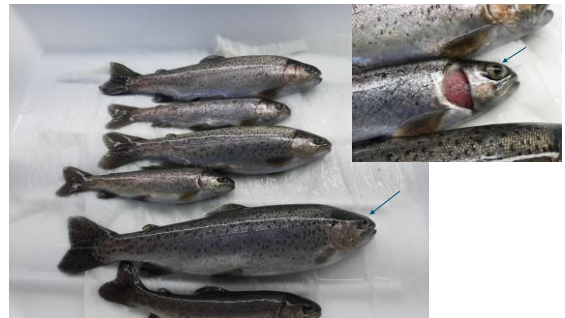
### Autopsy at WBVR, Lelystad

- Lethargy, paleness, light exophthalmos
  - Some Glossatella on the skin, not on the gills
  - Gills pale and congested with excess of slime, and with thickened gill tops, and necrosis
  - Liver pale-yellowish, gut content also yellowish
  - No muscular hemorrhages, but rather acute mortality
- => Veterinary Authority was contacted with a suspicion for notifiable fish diseases



3

### Clinics



4

### Autopsy and sampling

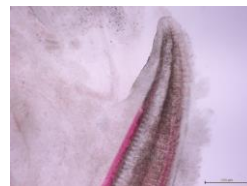


5

### Gills



- Congested, excess of slime, thickened gill tops, necrosis



6

Autopsy: internal organs



7

WBVR: Virus isolation, qPCRs, and bacteriology

- 10% organ suspension => on FHM and BF-2 cells (6-well plates) at 15°C => CPE after 3 days...
- Second passage: no further replication in both cell lines
- qPCR assays for VHSV, IHNV, IPNV, EHN, SAV and ISAV => all negative
- Bacteriology: *Flavobacterium* species isolated on TYES from skin and internal organs
- Still suspected for virus => [EURL contacted for help](#)



8

EURL Fish Diseases: Virology:

- 5 samples: 10% organ suspension, and (non-/filtered) supernatant of 1st passage at FHM and BF-2 cells
  - Cell culture at BF-2, EPC, CHSE, and FHM cells (24 well plates), for 2x7 days at 15°C => **no CPE**
  - qPCR assays for VHSV, IHNV, IPNV, RANAVirus, Piscine Orthoreovirus, PRV-1, PRV-3, and a conventional PCR for OMV
- => **Only Piscine Orthoreovirus** and the **PRV-3** qPCRs **positive**  
=> Detected in organ suspension and in FHM supernatant, and suspected for in BF-2 supernatant  
=> Fish farm culled and disinfected the pond



9

Conclusions

- First case of **Piscine Orthoreovirus genotype 3** in the Netherlands in July 2022.
  - It was combined with **flavobacteriosis**.
  - PRV-3 diagnosed by EURL by 2 qPCRs.
  - In Denmark PRV-3 is involved in cases of increased mortality often as part of a multi-factorial disease, like in this casus.
- => Atypical CPE seen, which cell culture to choose?  
=> Which other European countries have detected PRV-3?  
=> Should we implement the qPCRs for this infection?  
=> Enough to discover, as ever!



10



10

Thanks to our WBVR fish disease team:  
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11

Thank you for your attention

Questions?

*The girl with the pearl,*  
by  
Johannes Vermeer, 1665

<https://meesterwerkenaanjemeur.nl/meisje-met-de-pearl-vermeer-muurcirkel>



[olga.haenen@wur.nl](mailto:olga.haenen@wur.nl)

12