



# Viral genomic surveillance – supporting disease control in Atlantic salmon aquaculture

Bertie Knight (2<sup>nd</sup> year PhD student)

Trinations – 25/04/24

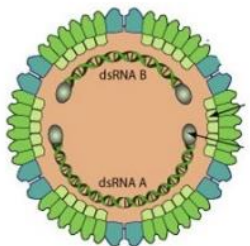


**Sustainable Aquaculture  
Innovation Centre**

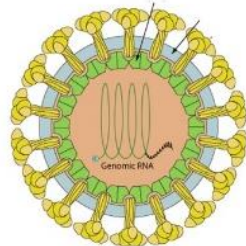


# How can viral genomic surveillance aid infection control?

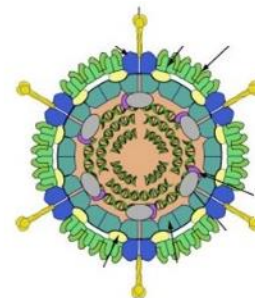
- Genetic diversity
  - Identify **emergence** of new viral subtypes
    - Vaccine updates
  - Understand **spatial distribution** of viral subtypes
    - Targeted control efforts
- Outbreak transmission scenarios
  - **Map transmission** of viral lineages



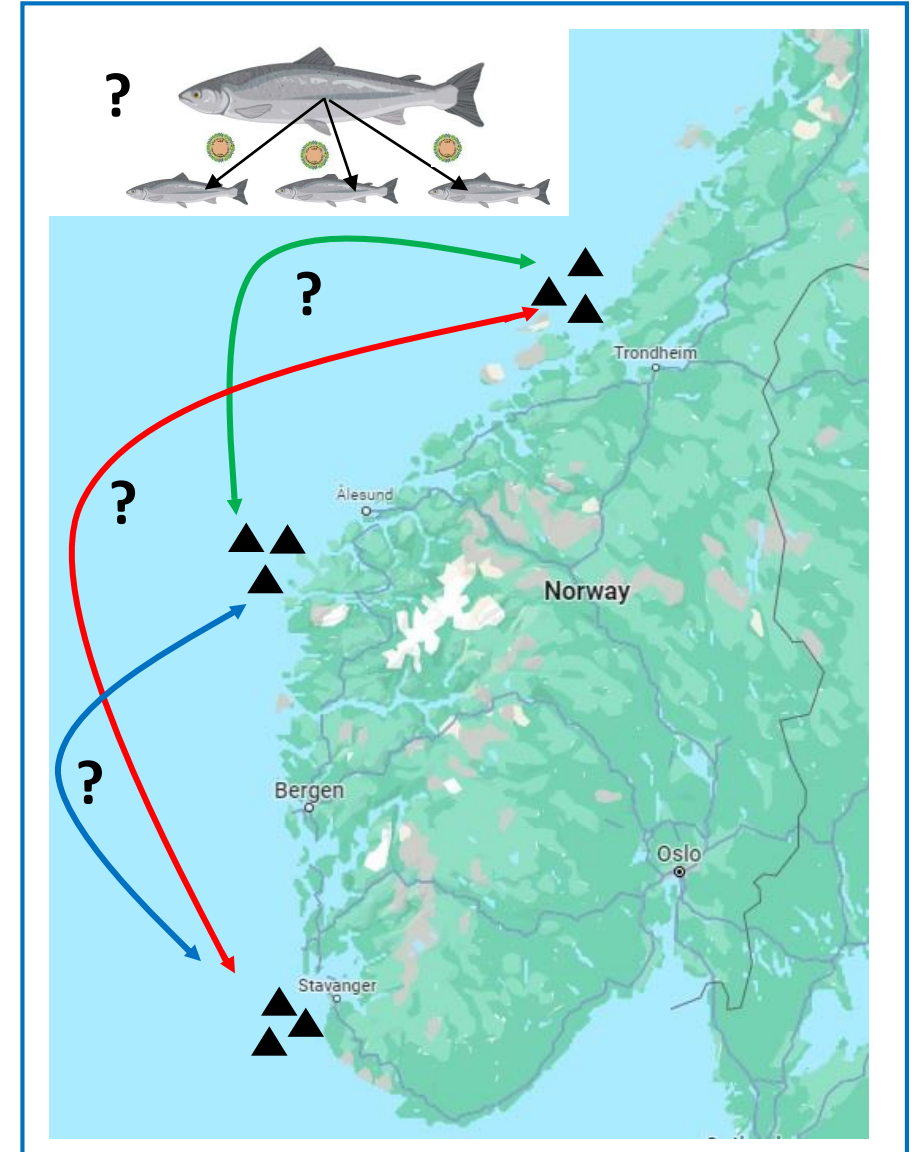
Infectious Pancreatic Necrosis Virus (IPNV)



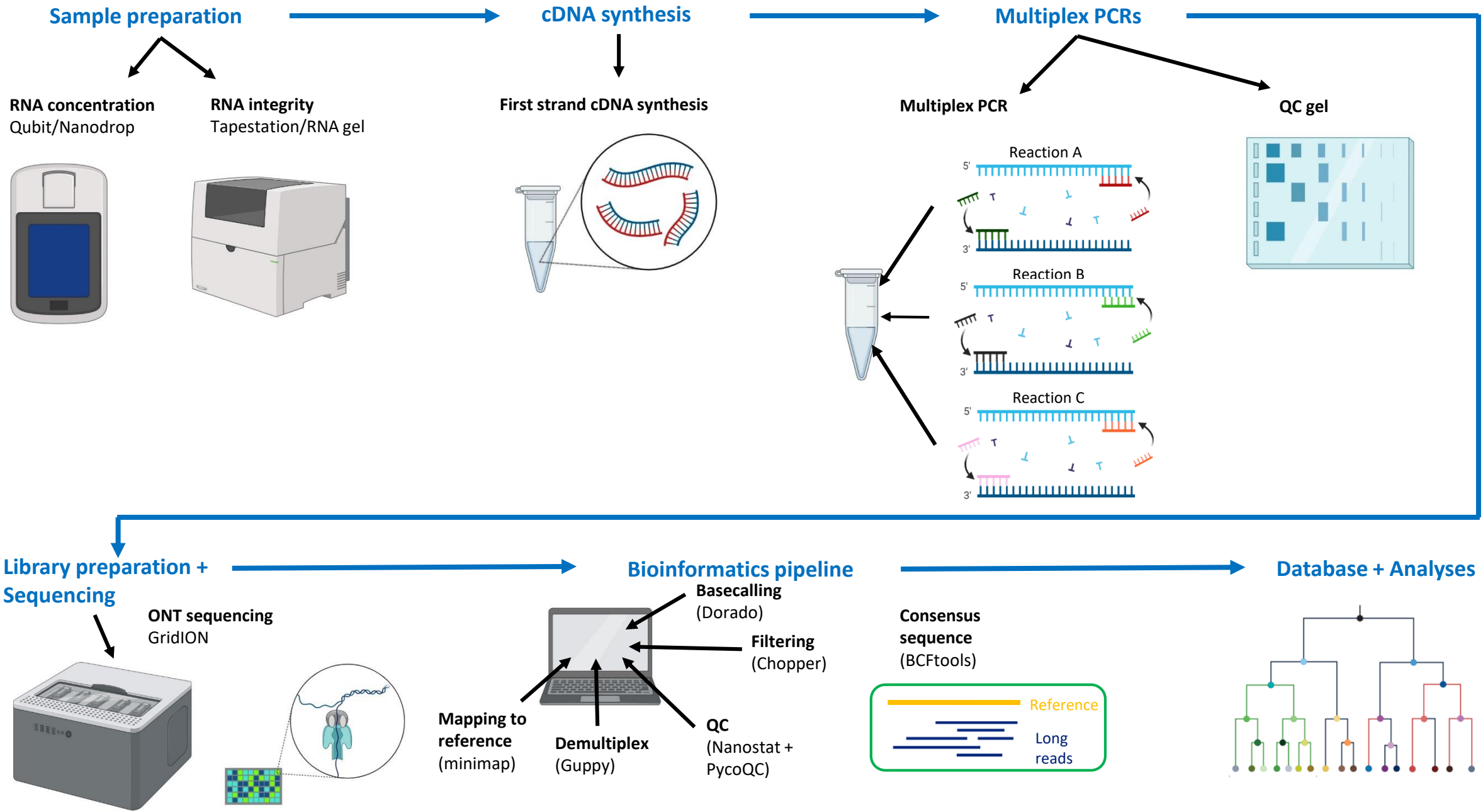
Salmonid alphavirus (SAV)



Piscine Orthoreovirus (PRV)



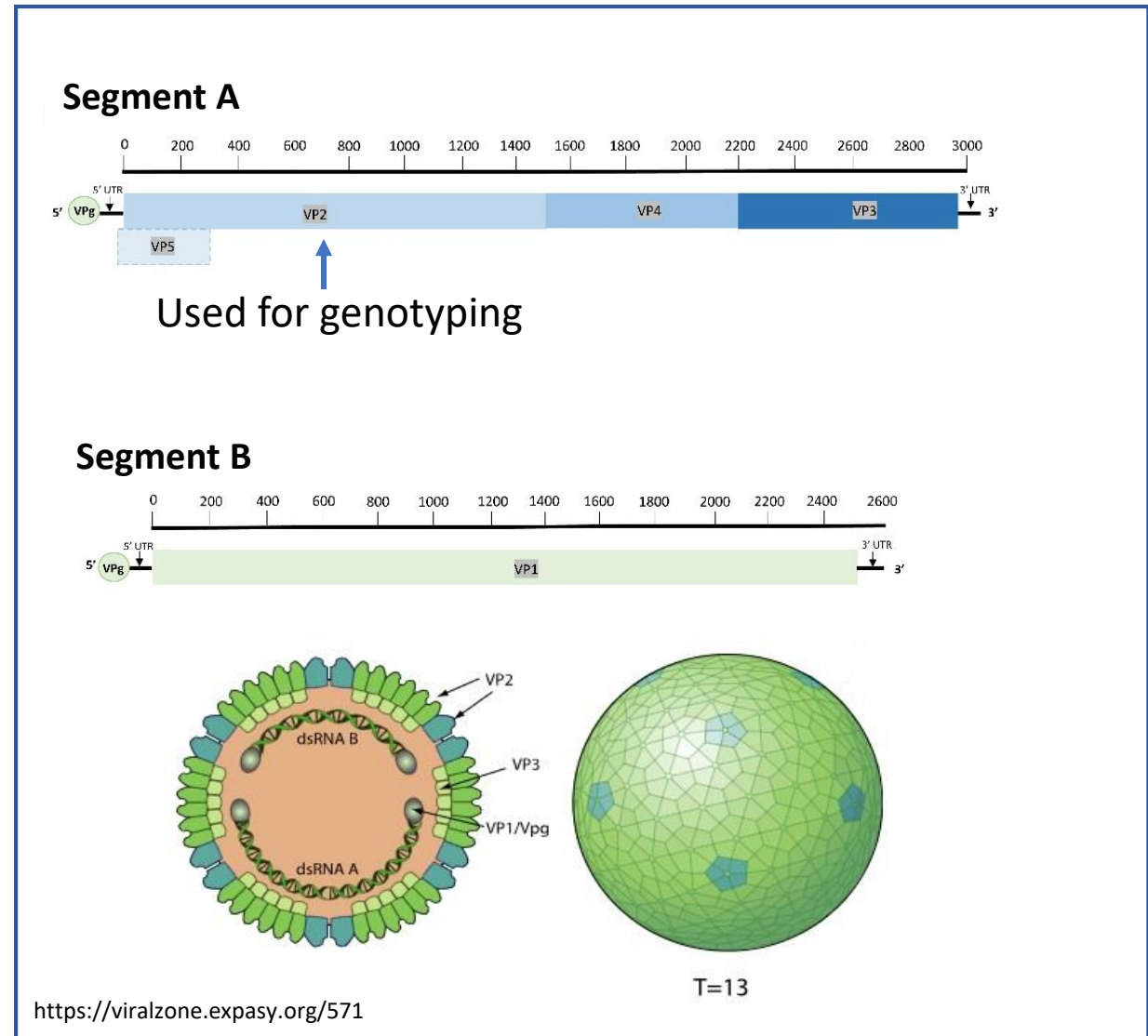
# Project workflow: sample to whole genome sequence



Origins of new Infectious  
Pancreatic Necrosis Virus (IPNV)  
variant of concern

# Infectious Pancreatic Necrosis Virus (IPNV)

- Family: Birnaviridae
- Bisegmented dsRNA (~6kb genome)
- Pancreas and liver necrosis
- Horizontal and possibly vertical transmission
- Phylogenetic relationship of VP2 used for genotyping: G1-G8
  - Scotland: G1 and G5
  - Norway: G5
- QTL for IPNV resistance (Houston *et al.* 2008)



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**GENETICS**  
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Genetics, 2015 Aug, 200(4): 1313–1326  
Published online 2015 Jun 2. doi: [10.1534/genetics.115.175406](https://doi.org/10.1534/genetics.115.175406)

PMCID: PMC4574245  
PMID: 26041276

Epithelial Cadherin Determines Resistance to Infectious Pancreatic Necrosis Virus in Atlantic Salmon

Thomas Moen,<sup>1,†</sup> Jacob Torgersen,<sup>†</sup> Nina Santi,<sup>†</sup> William S. Davidson,<sup>†</sup> Matthew Baranski,<sup>‡</sup> Jørgen Ødegård,<sup>†</sup> Sissel Kjøglum,<sup>†</sup> Bente Velle,<sup>§</sup> Matthew Kent,<sup>§</sup> Krzysztof P. Lubieniecki,<sup>†</sup> Eivind Isdal,<sup>†\*</sup> and Sigbjørn Lien<sup>§</sup>

The nedd-8 activating enzyme gene underlies genetic resistance to infectious pancreatic necrosis virus in Atlantic salmon

Ion Pavelin,<sup>1,‡</sup> Ye Hwa Jin,<sup>1,‡</sup> Remi L. Gratacap,<sup>2,§</sup> John B. Toqqart,<sup>3,§</sup> Alastair Hamilton,<sup>4,§</sup> David W. Verner-Jeffreys,<sup>4,§</sup> Richard K. Paley,<sup>4,§</sup> Carl-Johan Rubin,<sup>4,§</sup> Stephen C. Bishop,<sup>4,§</sup> James E. Bron,<sup>5,§</sup> Diego Robledo,<sup>5,§</sup> Ross D. Houston,<sup>6,§</sup>

# IPN disease in vaccinated/QTL Atlantic salmon



Feb 2021



Article

**Isolation of a New Infectious Pancreatic Necrosis Virus (IPNV) Variant from a Fish Farm in Scotland**

Jessica Benkaroun <sup>1</sup>, Katherine Fiona Muir <sup>1</sup>, Rosa Allshire <sup>1</sup>, Cüneyt Tamer <sup>2</sup> and Manfred Weidmann <sup>1,3,\*</sup>



Oct 2021

**Identification of a New Infectious Pancreatic Necrosis Virus (IPNV) Variant in Atlantic Salmon (*Salmo salar* L.) that can Cause High Mortality Even in Genetically Resistant Fish**



Borghild Hillestad, Stein Johannessen, Geir Olav Melingen and Hooman K. Moghadam \*



Oct 2022



Article

**Isolation of a New Infectious Pancreatic Necrosis Virus (IPNV) Variant from Genetically Resistant Farmed Atlantic Salmon (*Salmo salar*) during 2021–2022**

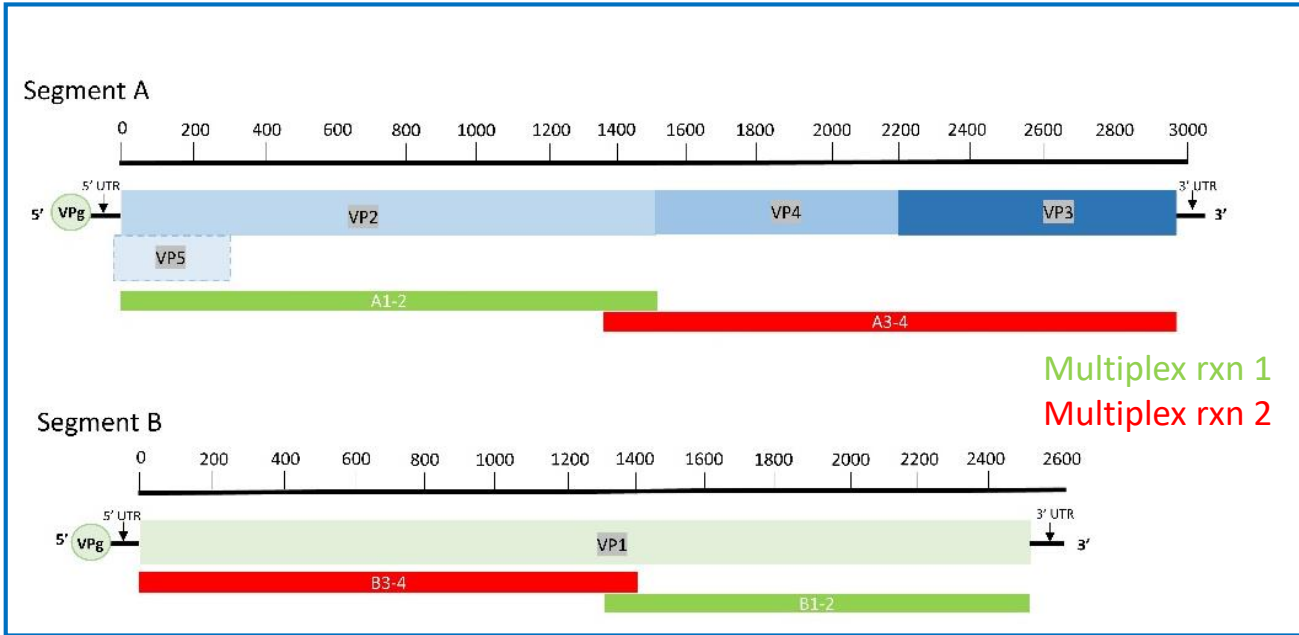
Marcos Godoy <sup>1,2,\*</sup>, Molly J. T. Kibenge <sup>3</sup>, Marco Montes de Oca <sup>2</sup>, Juan Pablo Pontigo <sup>1</sup>, Yoandy Coca <sup>4</sup>, Diego Caro <sup>2</sup>, Karina Kusch <sup>2</sup>, Rudy Suarez <sup>5</sup>, Ian Burbulis <sup>6</sup> and Frederick S. B. Kibenge <sup>3</sup>

Reports from industry suggesting increasing IPNV outbreaks in QTL/vaccinated fish

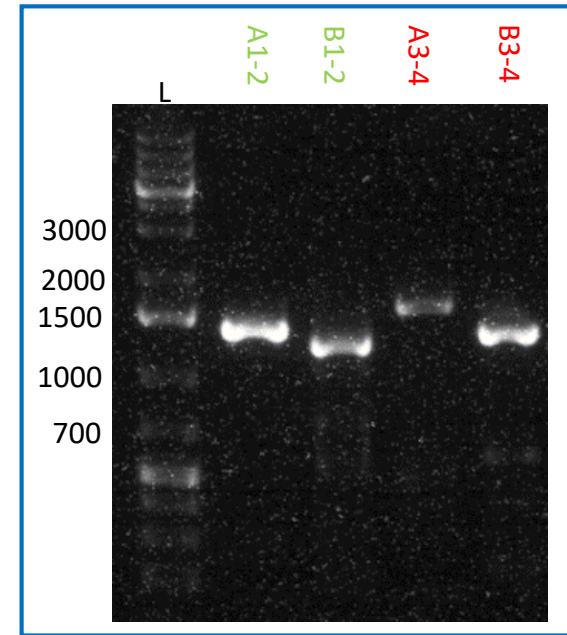
Novel G5 variant

# IPNV amplicon-based multiplex approach

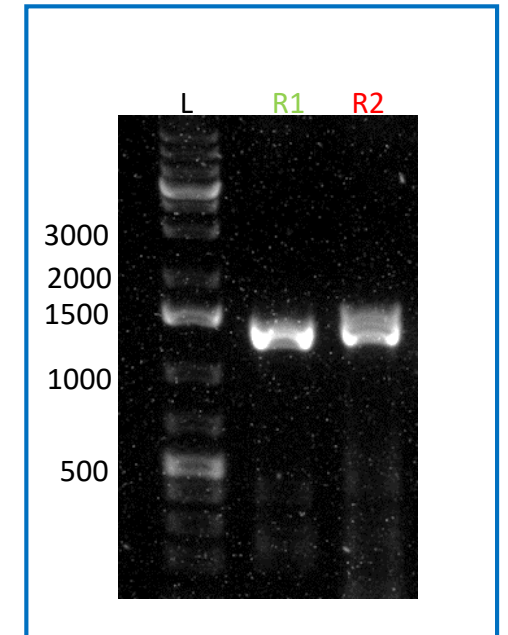
IPNV multiplex scheme



Individual primers

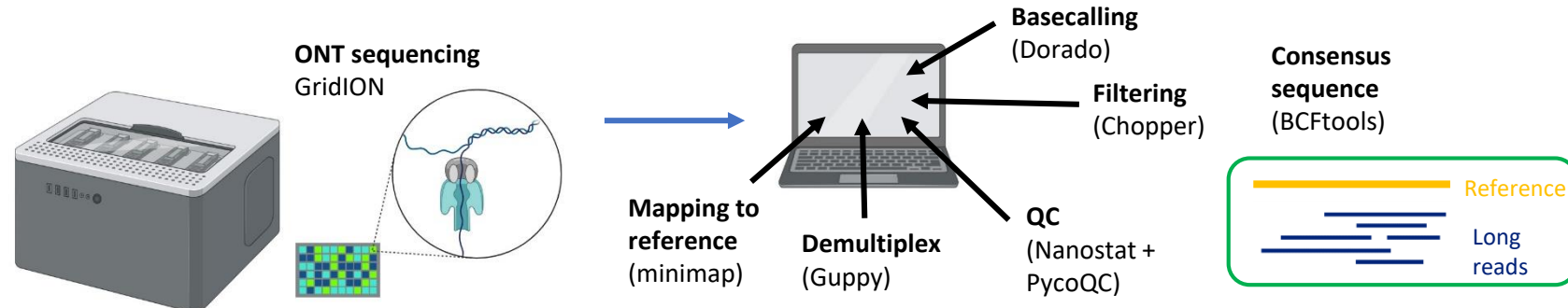


Multiplexed primers

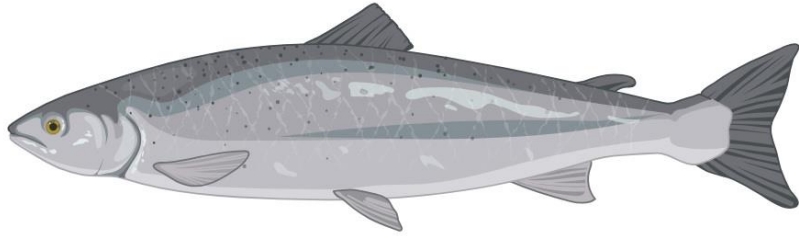


Primers targeted to "Novel" G5 but can also capture "Classic" G5

Sequenced 69 whole genome IPNV sequences



# Maximum-likelihood phylogram of whole IPNV genome nucleotide sequences



NG5: Rapid evolution

Novel G5 has become predominant strain in Norway

"Novel" G5 (NG5)

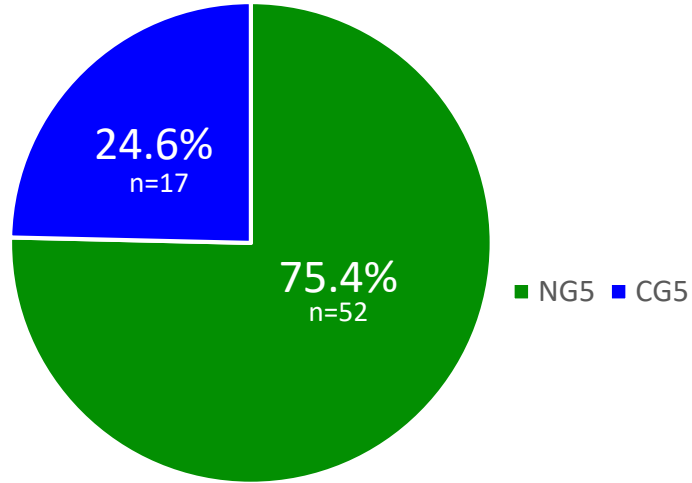
"Classic" G5 (CG5)

**177 whole genome sequences:**

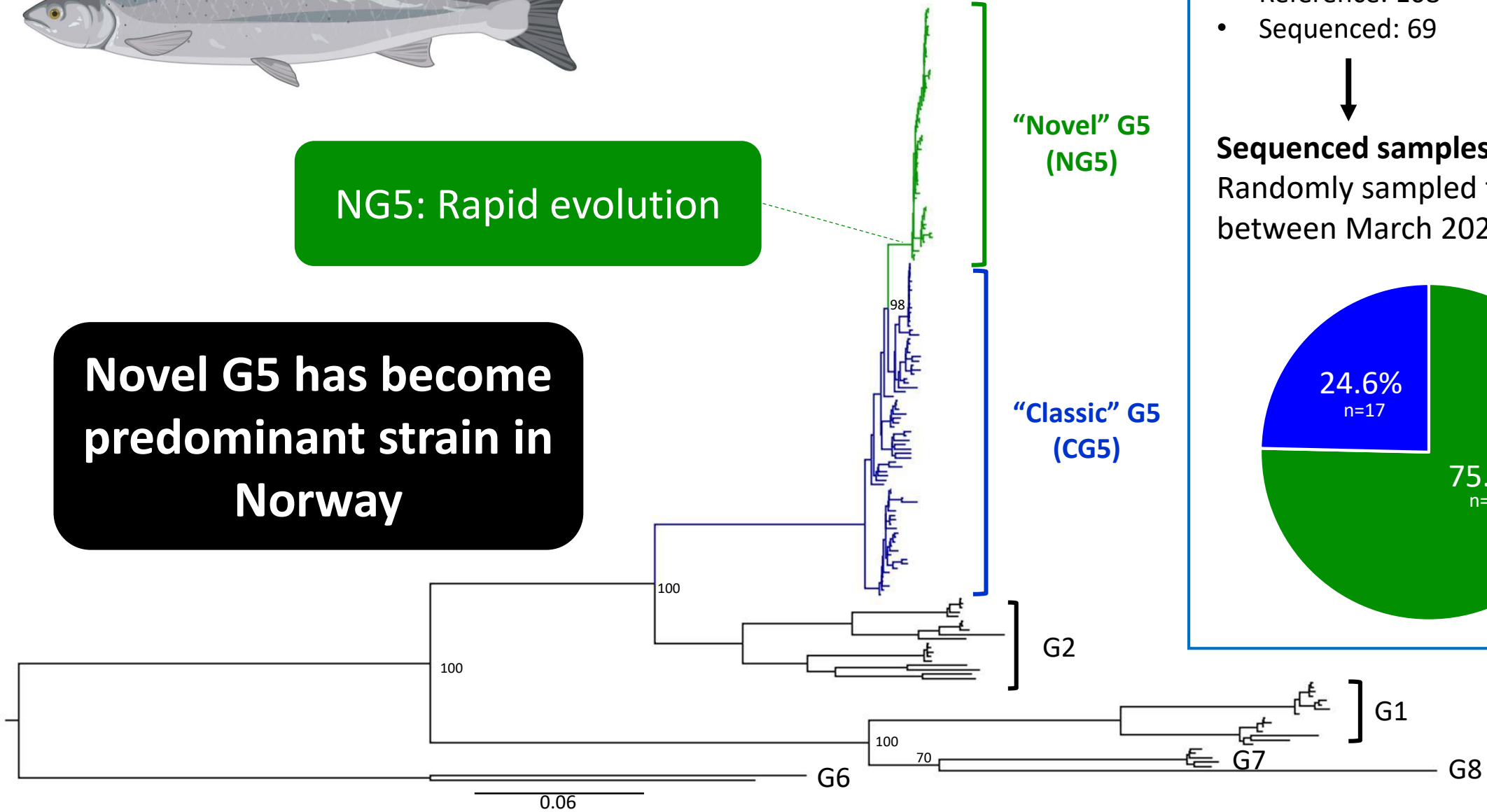
- Reference: 108
- Sequenced: 69

↓

**Sequenced samples (n = 69):**  
Randomly sampled from outbreaks between March 2022 and June 2023



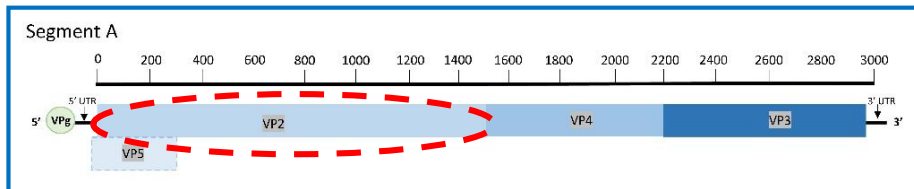
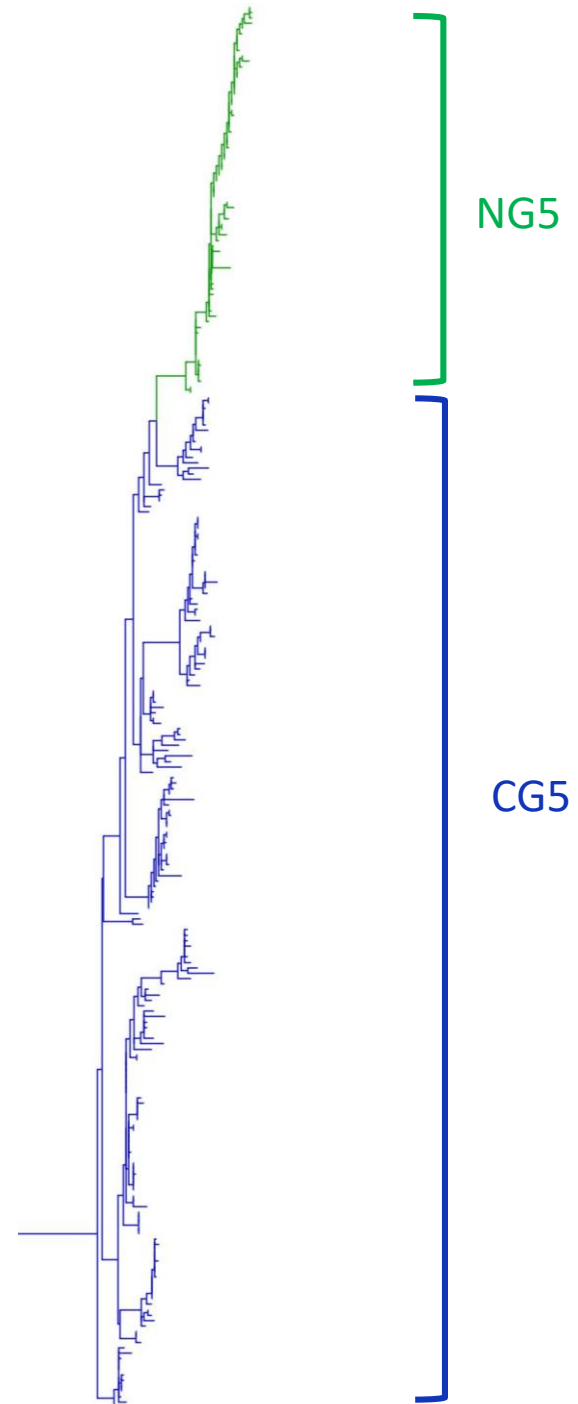
| Strain | Percentage | Count (n) |
|--------|------------|-----------|
| NG5    | 75.4%      | 52        |
| CG5    | 24.6%      | 17        |



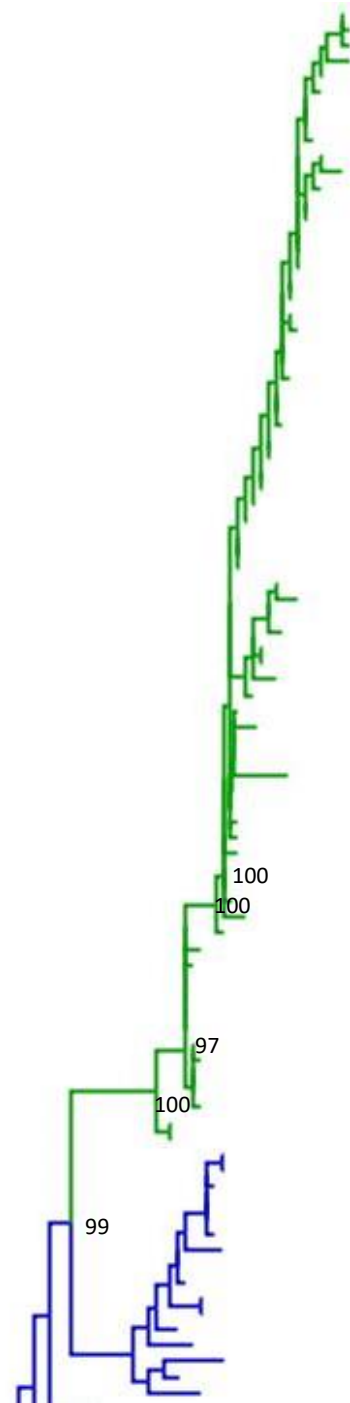
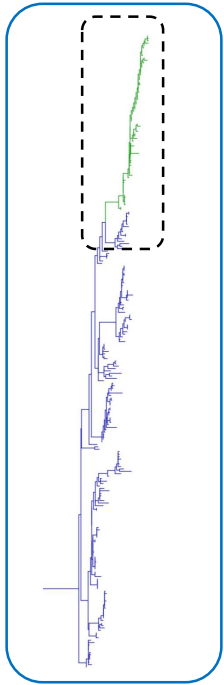


# Maximum-likelihood phylogram of VP2 nucleotide sequences

369 VP2 sequences (300 reference + 69 sequenced samples)



# Maximum-likelihood phylogram of VP2 nucleotide sequences



Sequenced in this study  
(n=52) (March 2022 –  
June 2023)

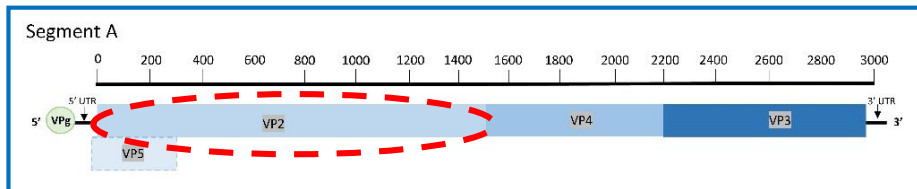


Novel G5 refs  
Scotland (2018)  
Norway (2019)

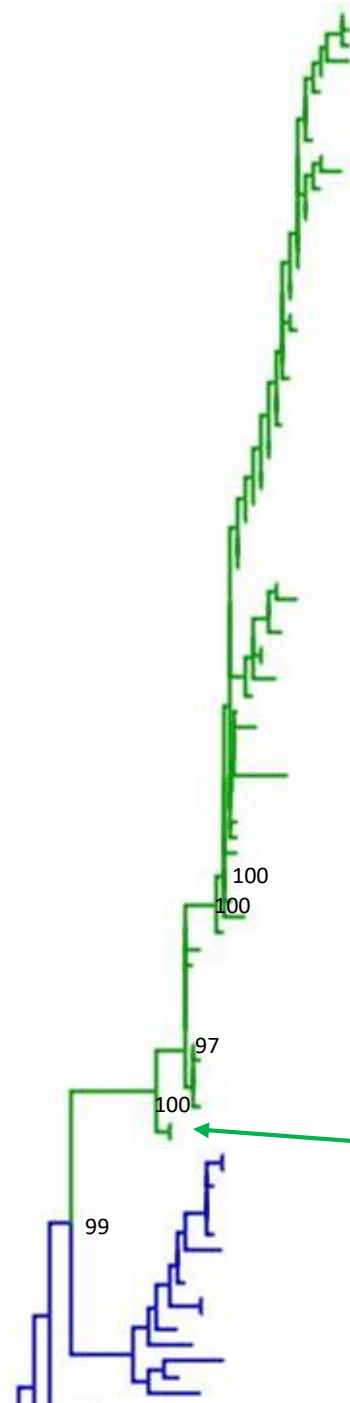
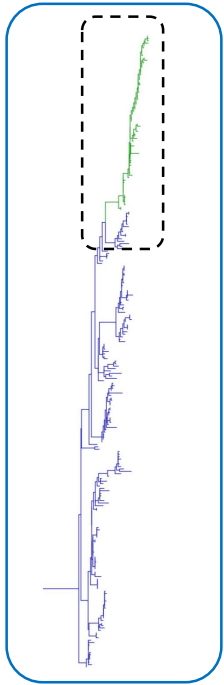


NG5

CG5



# Maximum-likelihood phylogram of VP2 nucleotide sequences



Sequenced in this study  
(n=52) (March 2022 –  
June 2023)



Novel G5 refs  
Scotland (2018)  
Norway (2019)



**Previously unidentified NG5**

Norwegian references  
(2012 – 2014)



**Previously unidentified NG5**

Italian references  
(2010 – 2015)

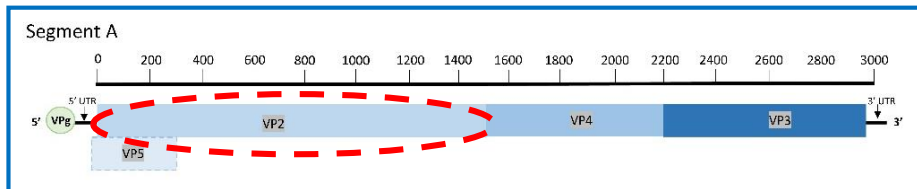


**Previously unidentified NG5**  
Norwegian references  
(2004)



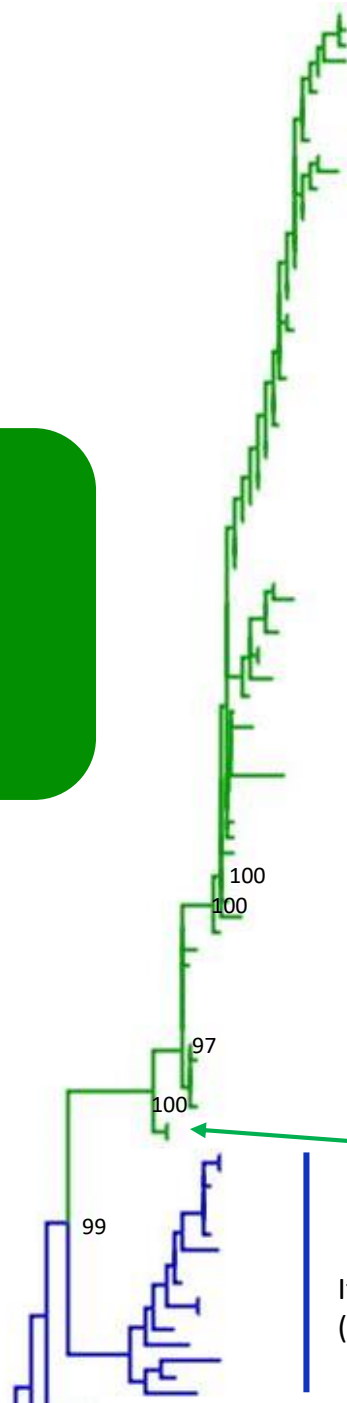
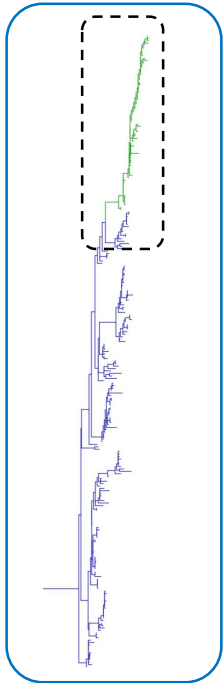
NG5

CG5



# Maximum-likelihood phylogram of VP2 nucleotide sequences

**Hypothesis: Norwegian NG5 in Atlantic salmon originated in freshwater salmonid aquaculture**



Sequenced in this study (n=52) (March 2022 – June 2023)



Novel G5 refs  
Scotland (2018)  
Norway (2019)



**Previously unidentified NG5**

Norwegian references (2012 – 2014)



**Previously unidentified NG5**

Italian references (2010 – 2015)



**Previously unidentified NG5**  
Norwegian references (2004)

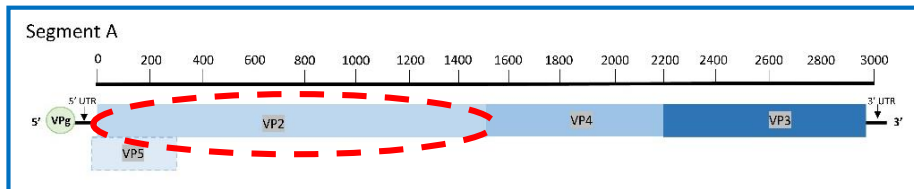


Italian references (1978 – 2015)



NG5

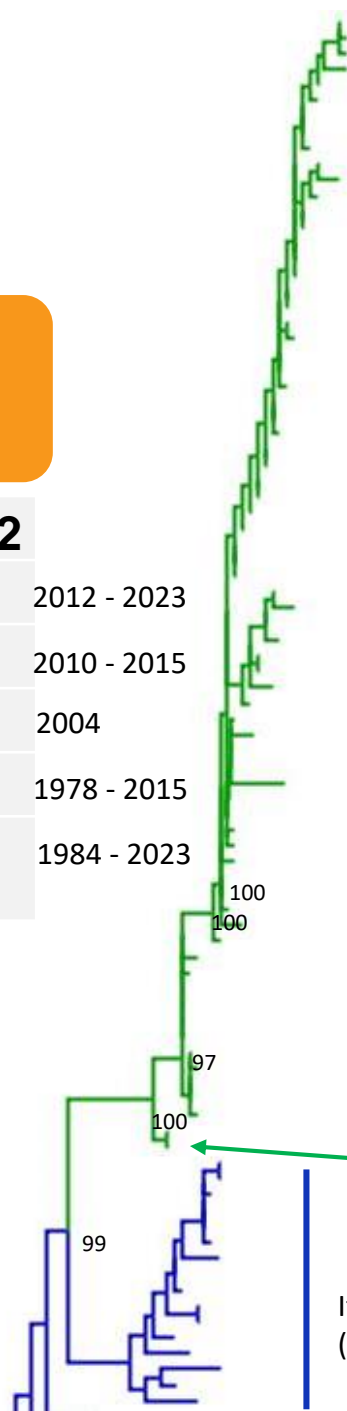
CG5



# Maximum-likelihood phylogram of VP2 nucleotide sequences

Comparison of amino acid sequence in the VP2 hypervariable region (239-257 + 271-284)

|                                 | 245 | 248 | 252 | 255 | 257 | 282 |             |
|---------------------------------|-----|-----|-----|-----|-----|-----|-------------|
| Norwegian/Scottish NG5          | G   | R   | D   | T   | H   | T   | 2012 - 2023 |
| Italian NG5                     | G   | G   | D   | T   | H   | N   | 2010 - 2015 |
| Norwegian NG5                   | G   | G   | D   | T   | H   | N   | 2004        |
| Italian "classic" G5            | S   | A/E | N   | K   | H   | N   | 1978 - 2015 |
| Norwegian/Scottish "classic" G5 | S   | E   | N   | K   | D   | D   | 1984 - 2023 |






Sequenced in this study (n=52) (March 2022 – June 2023)

 n=51  n=1  *S. salar*

Novel G5 refs  
Scotland (2018)    *S. salar*  
Norway (2019)

Previously unidentified NG5  
Norwegian references (2012 – 2014)   *S. salar*

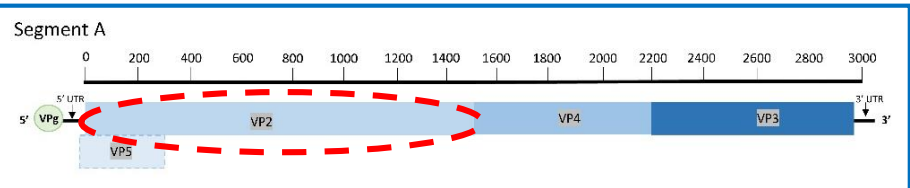
Previously unidentified NG5  
Italian references (2010 – 2015)   *O. mykiss*  *Salvelinus* spp.

Previously unidentified NG5  
Norwegian references (2004)   *S. salar*

Italian references (1978 – 2015)   *O. mykiss*

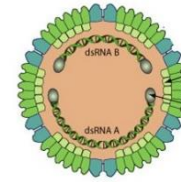
NG5

CG5

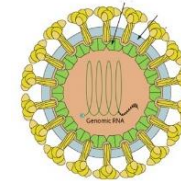


# Perspectives and future work

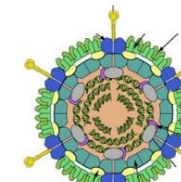
- Spatial-temporal reconstruction of transmission scenarios
  - Update viral molecular epidemiology
  - Characterise genetic diversity
- Sequencing assays are scalable
  - Hundreds of viral whole genome sequences for study



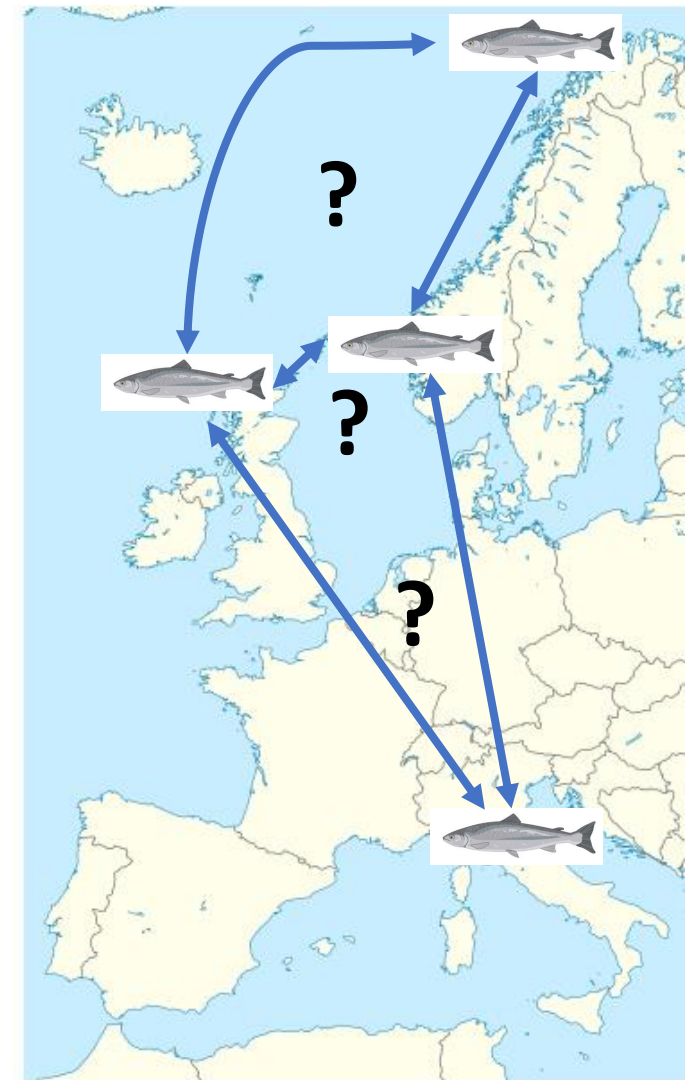
Infectious Pancreatic Necrosis Virus (IPNV)



Salmonid alphavirus (SAV)



Piscine Orthoreovirus (PRV)



Viral genomic data can support disease control management

# Acknowledgements

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Ane Sandtrø



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Jørgensen



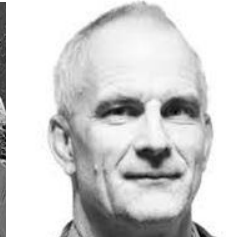
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Solheim



Chris Matthews



Marius Karlsen



Svein  
Alexandersen



Elise Hjelle

## Royal Veterinary College



Sarah Hill



Mingli Zhao



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Biological Sciences  
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