

Molecular diagnostics of viral diseases in Irish aquaculture

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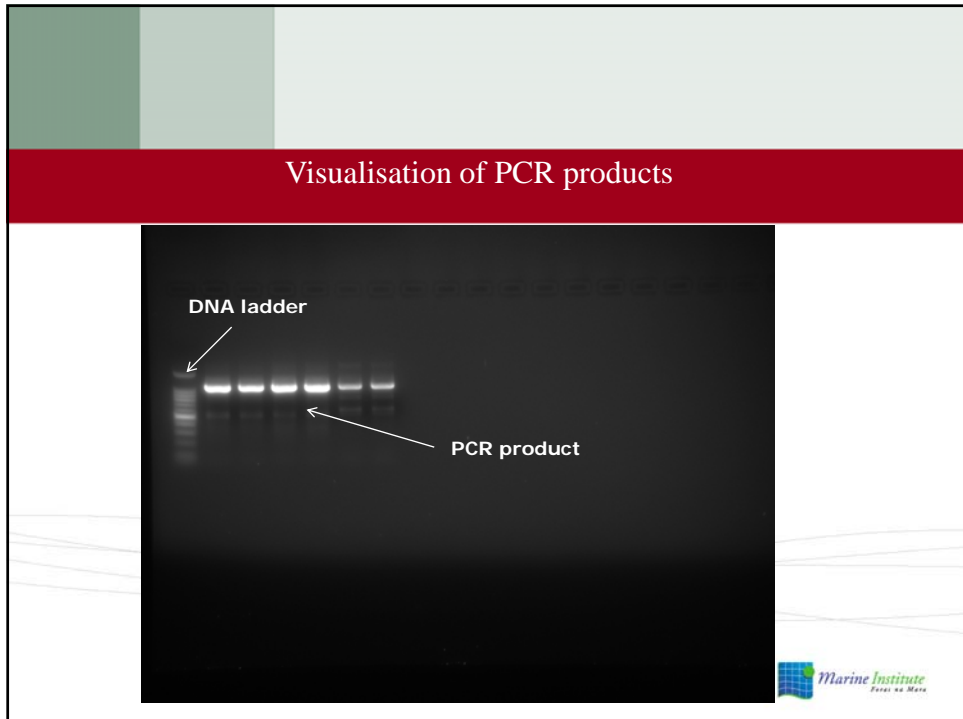
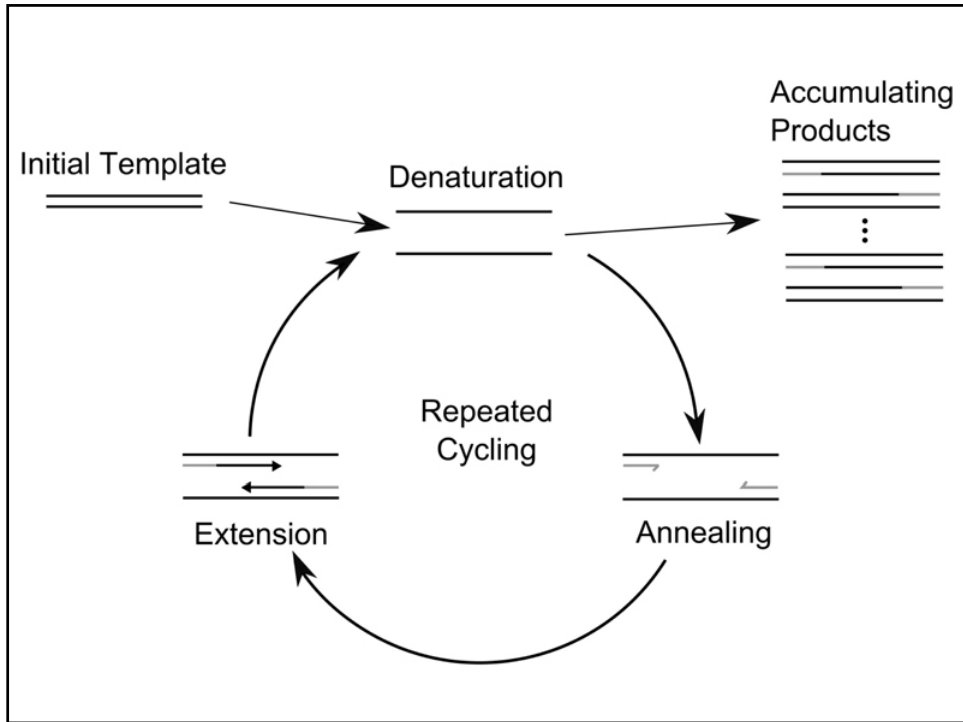
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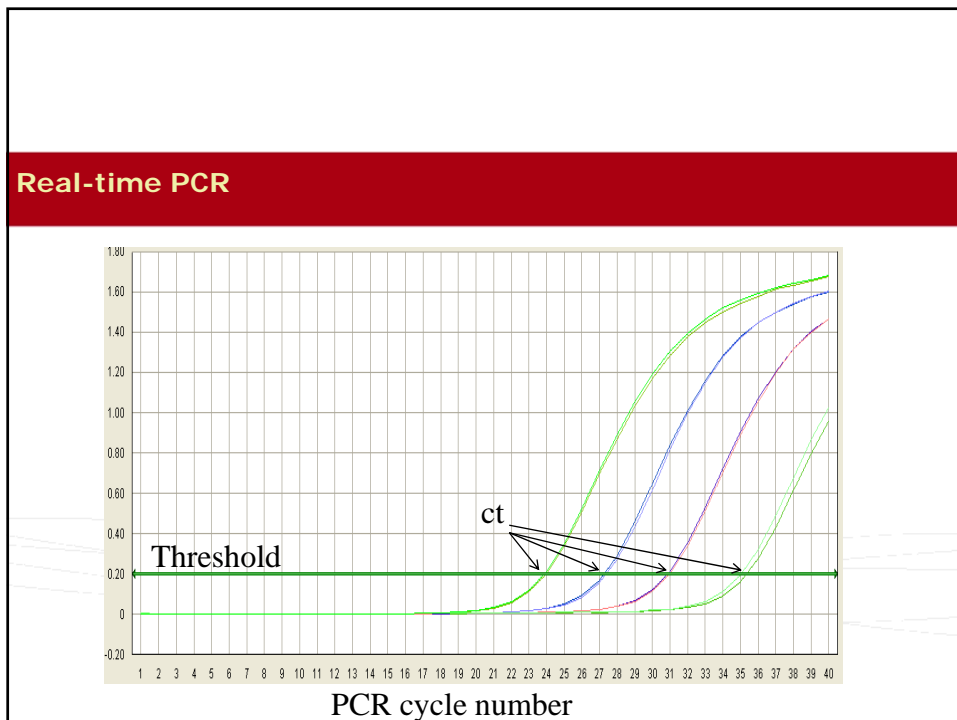
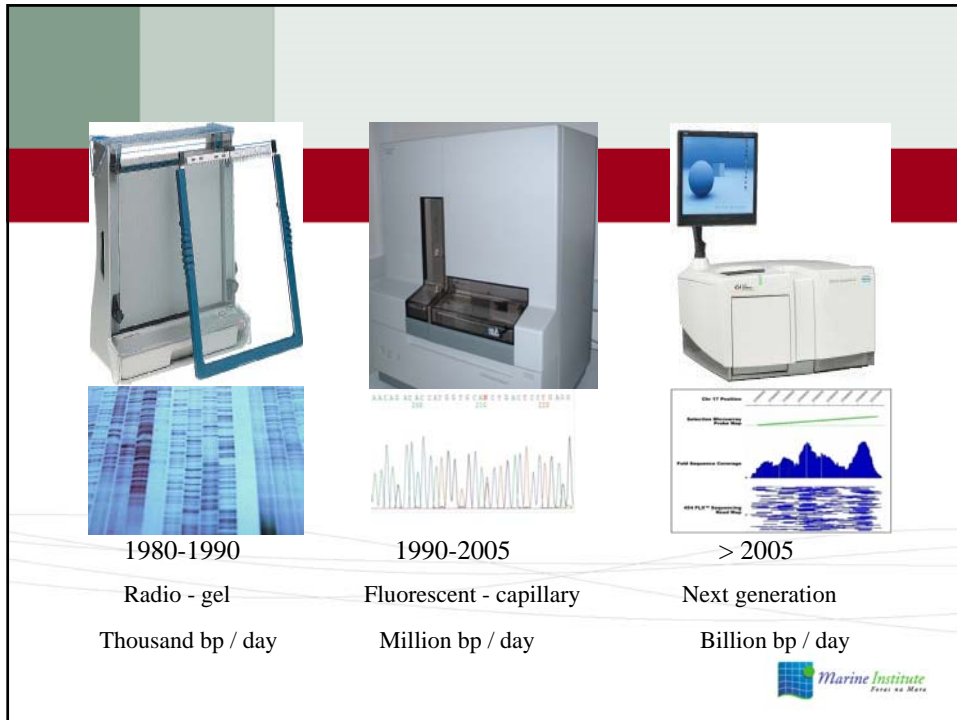



Background

- based on the amplification of the viral genome
- use pathogen specific primers
- sensitive e.g. real-time PCR
- quick turnaround and large sample volumes

- expensive
- results should be considered along with more classical diagnostic methods (histopathology, cell culture etc.)



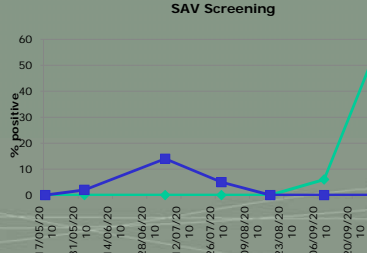





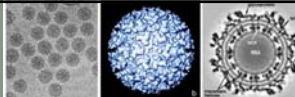
Using real-time PCR to screen for pancreas disease :

- non-lethal sampling (blood)
- typically 30 fish can be tested in one day
- detects initial infection stage

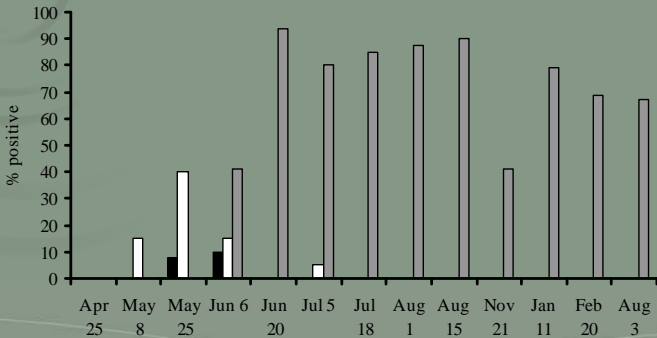
SAV Screening



Date	% positive
17/05/20	0
31/05/20	0
14/06/20	0
28/06/20	0
12/07/20	15
26/07/20	5
09/08/20	0
23/08/20	0
06/09/20	5
20/09/20	55





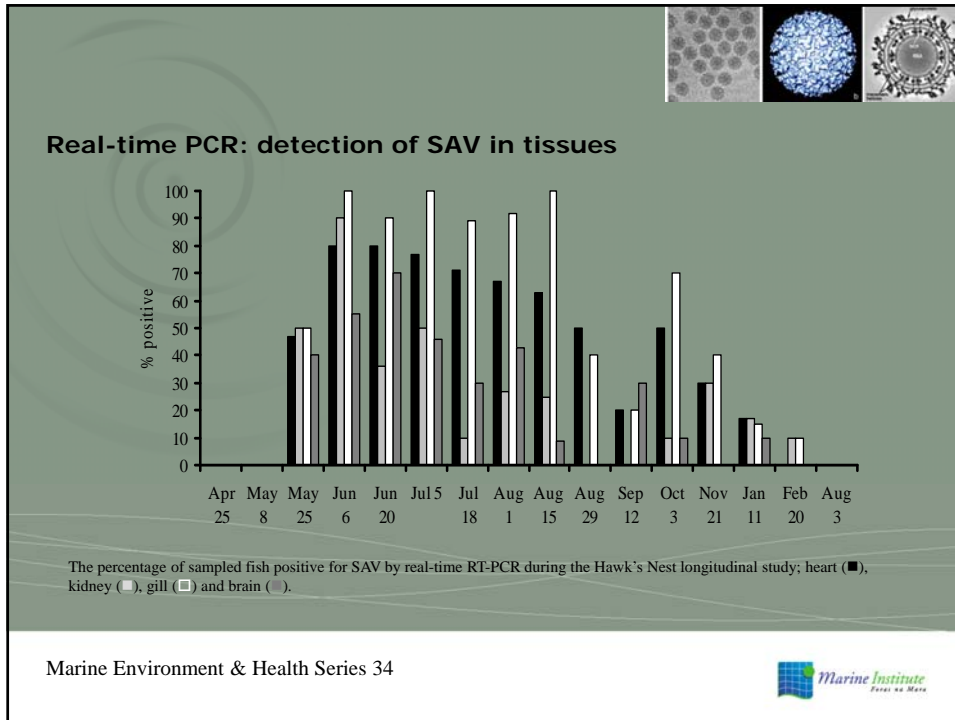
Real-time PCR v Serology



Date	Virus Isolation on Serum (%)	Real-time RT-PCR (%)	Antibody-positive Sentinel Fish (%)
Apr 25	0	0	0
May 8	0	15	0
May 25	10	40	0
Jun 6	12	15	42
Jun 20	0	0	95
Jul 5	0	5	80
Jul 18	0	0	85
Aug 1	0	0	90
Aug 15	0	0	92
Nov 21	0	0	42
Jan 11	0	0	80
Feb 20	0	0	70
Aug 3	0	0	68

The percentage of fish positive for SAV using virus isolation on serum (■), real-time RT-PCR (□) and the percentage of antibody-positive sentinel fish (■)





Longitudinal study overview

- Wk 4: SAV detected in blood by PCR
- Wk 6: SAV detected in blood by cell culture
 - First signs of disease by histopathology
 - SAV detected in fish tissues by PCR
- WK 8: First seropositive fish (anti-SAV antibodies detected)
- Wk 9/10: Mortalities increase
- Wk 16/17: Mortalities decreased, histopathology clear

Marine Environment & Health Series 34

Detection of SAV in wild fish:

2010 Snow et al reported the detection of salmonid alphavirus from tissue samples of dab (*Limanda limanda*) long rough dab (*Hippoglossoides platessoides*) and plaice (*Pleuronectes platessa*).

Could not culture the virus.

Virus was a subtype V.



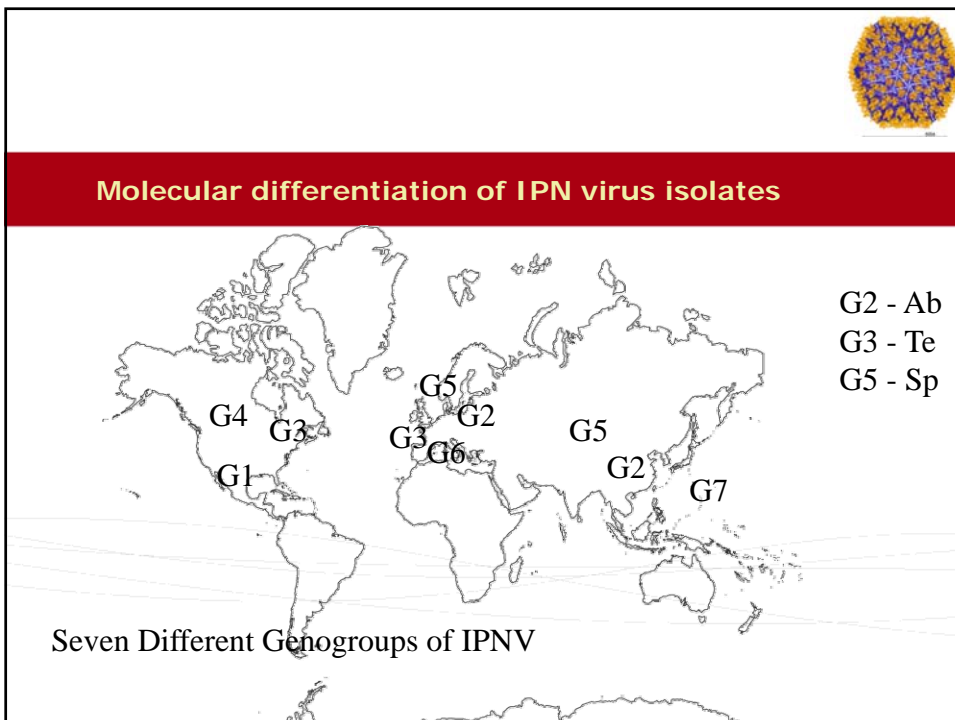
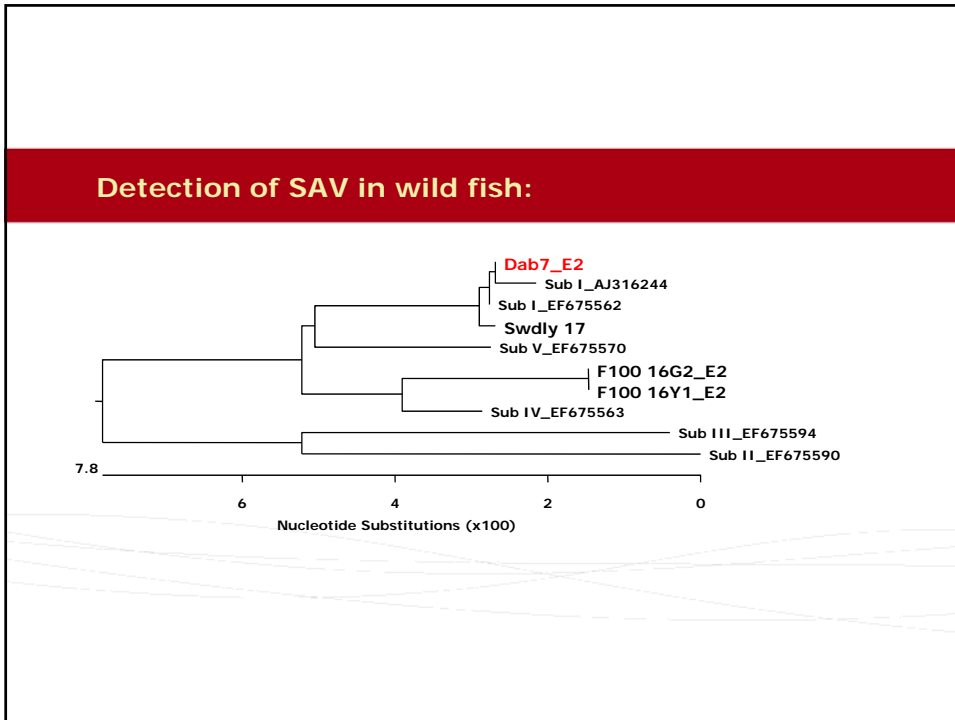
Detection of SAV in wild fish:

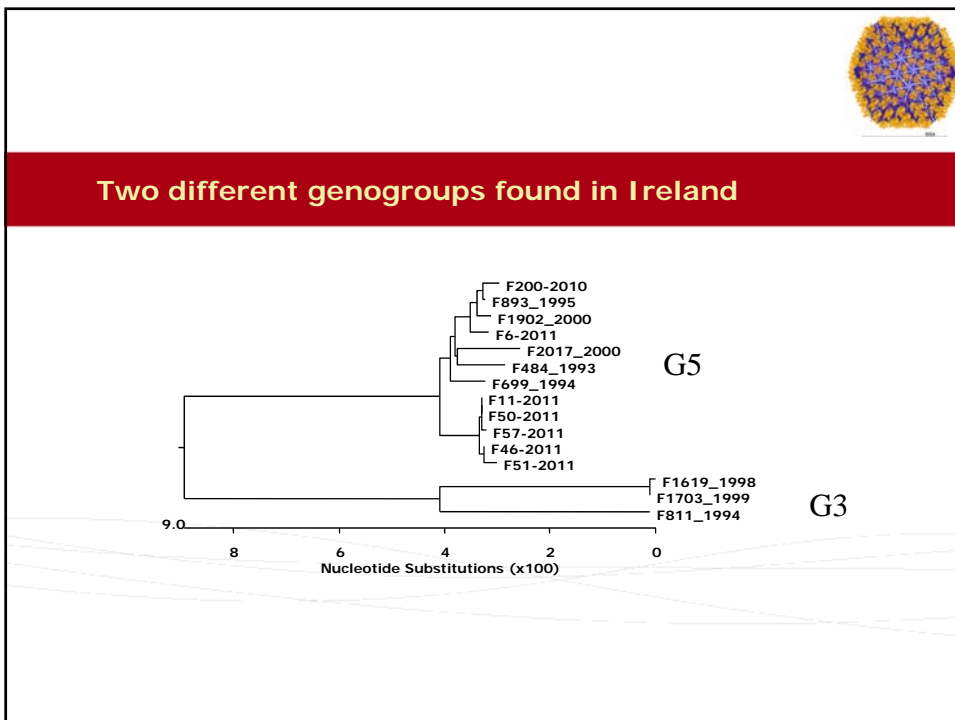
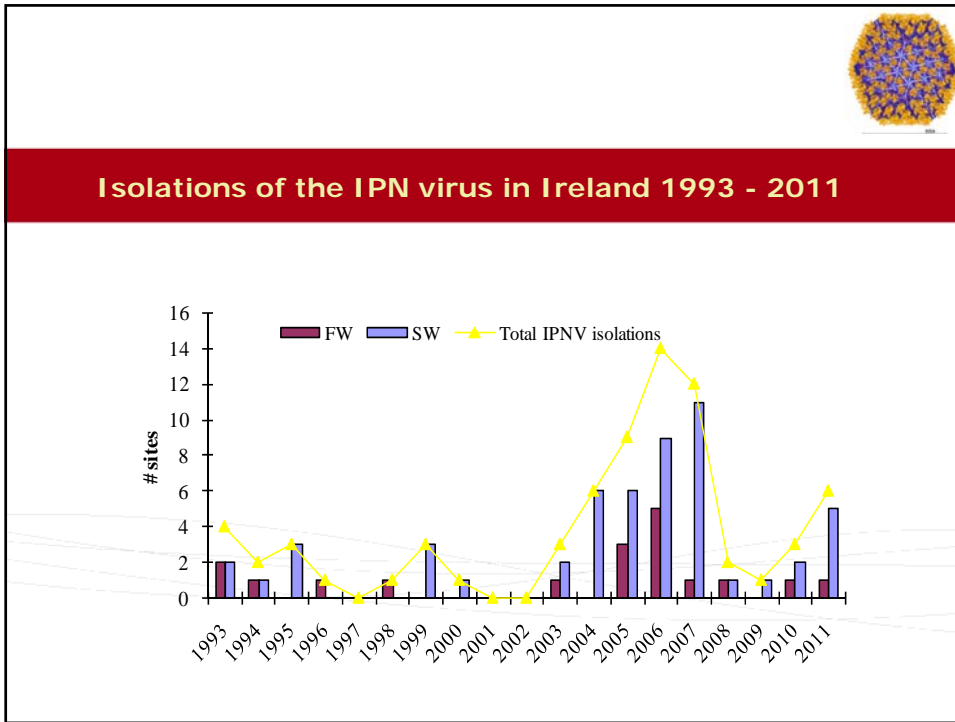
2 cruises in Irish Sea:

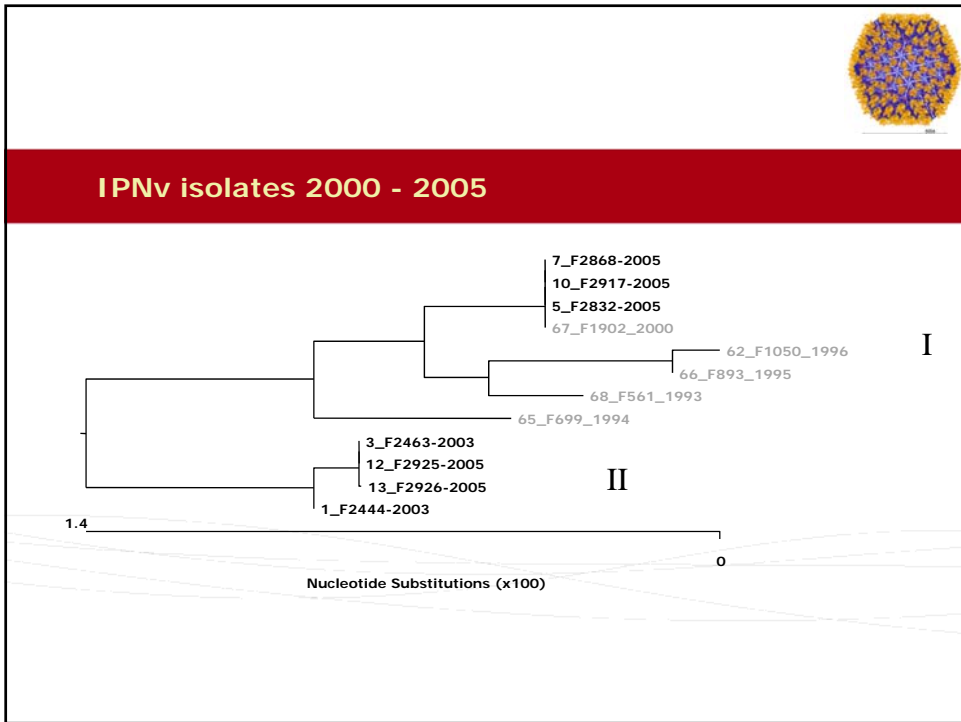
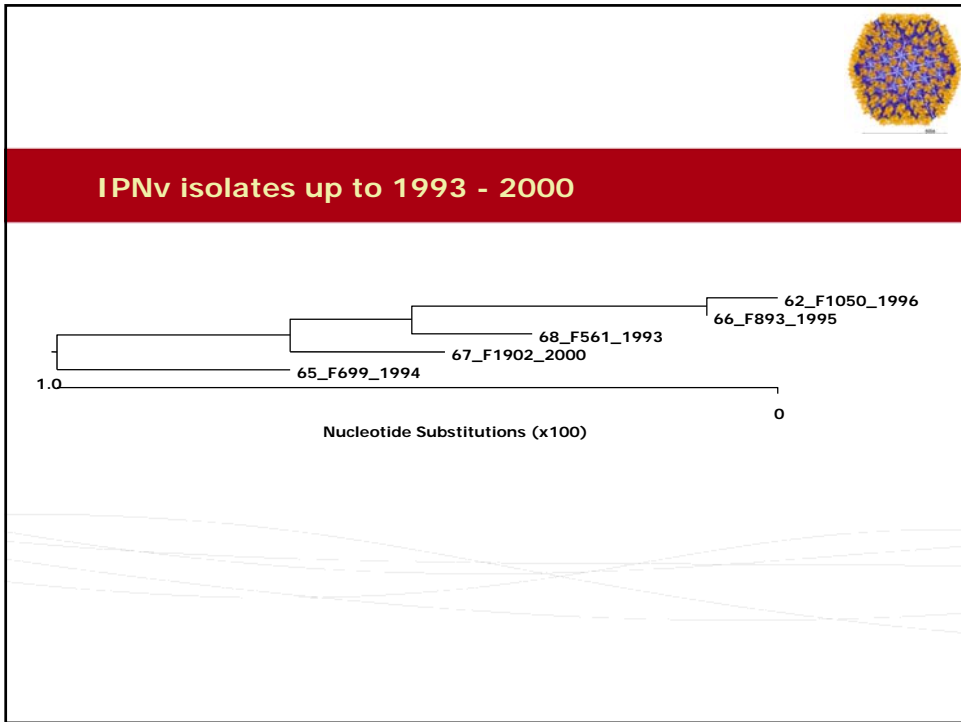
1. 100 dab/plaice tested (heart): 8 dab/5 plaice positive
2. 25 dab/plaice tested (gill): 2 dab positive

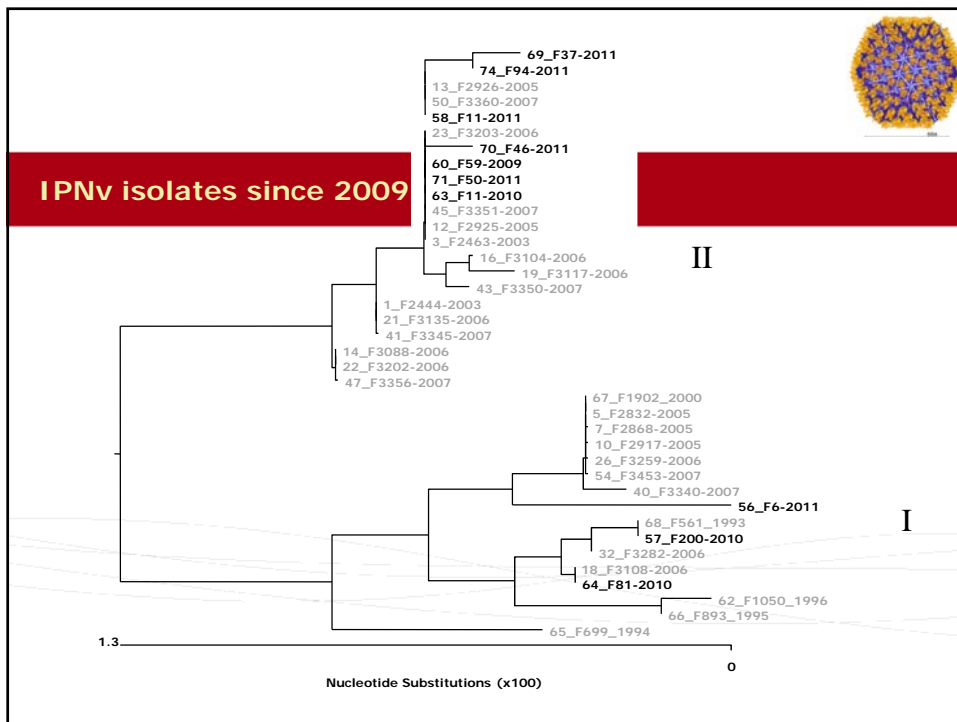
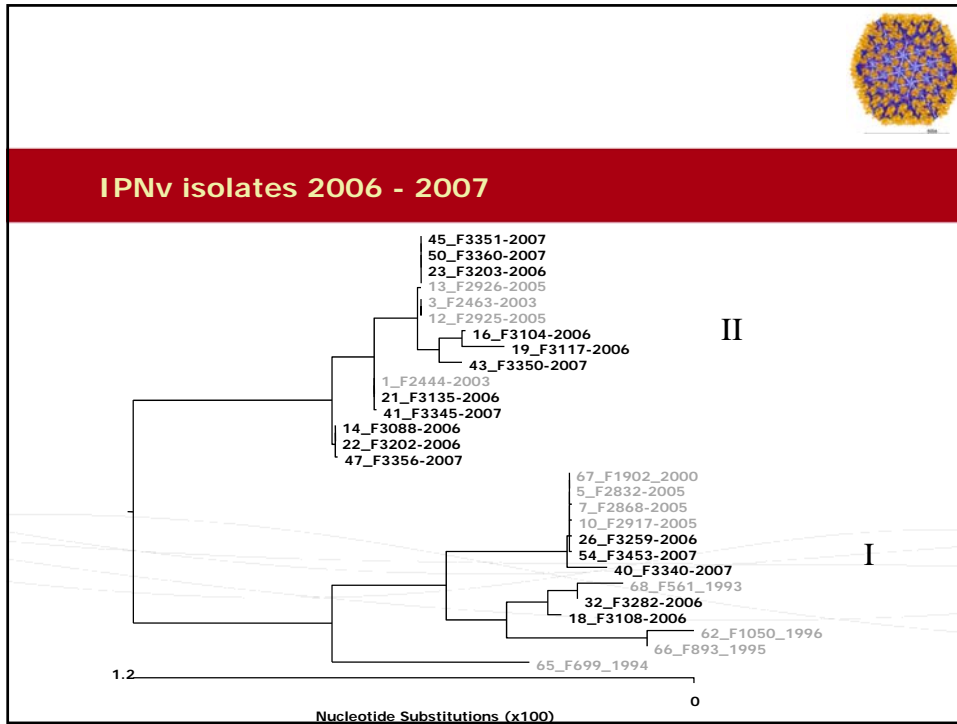
No fish farming in this area.

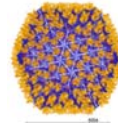
Are flatfish a host or have they somehow become infected?





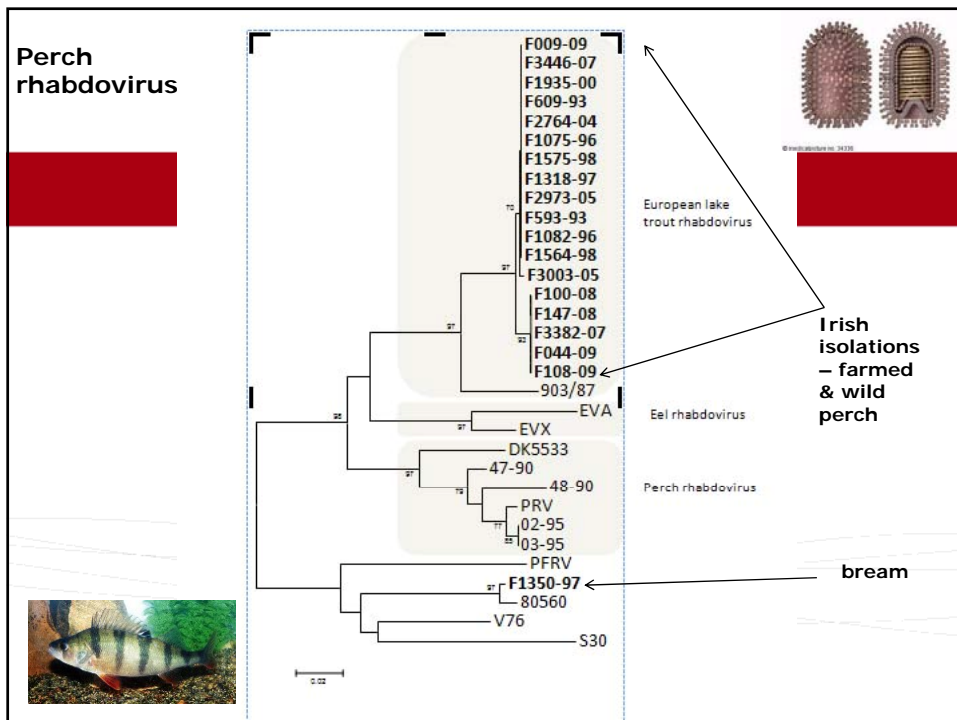






IPN conclusions

- Sub-group I has been present in the country since 1990's
- More virulent sub-group II first isolated in 2003 with the first officially reported clinical IPN outbreaks
- Sub-group II had been associated with imported fish
- Now appears to be spreading in marine sites where it has not previously been recorded
- Apart from broodstock and wild salmon (1) sub-group I isolates have not been found since 2007 – Outcompeted? Mutated?



My thanks to the staff of the MI Fish Health Unit

Questions?

