NoroRisk:

Developing a Risk Assessment Framework for Norovirus in Irish Oyster production Areas

DAFM FIRM Project: 14/F852
progress update

Dublin, 13th September 2016

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Overview

- Task 1. Ongoing monitoring results
- Task 2. Depuration experiments – set up and results
- Future work

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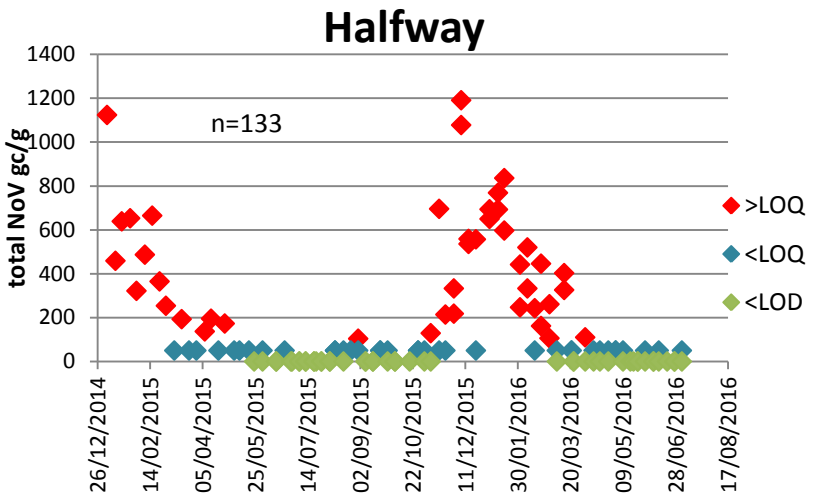
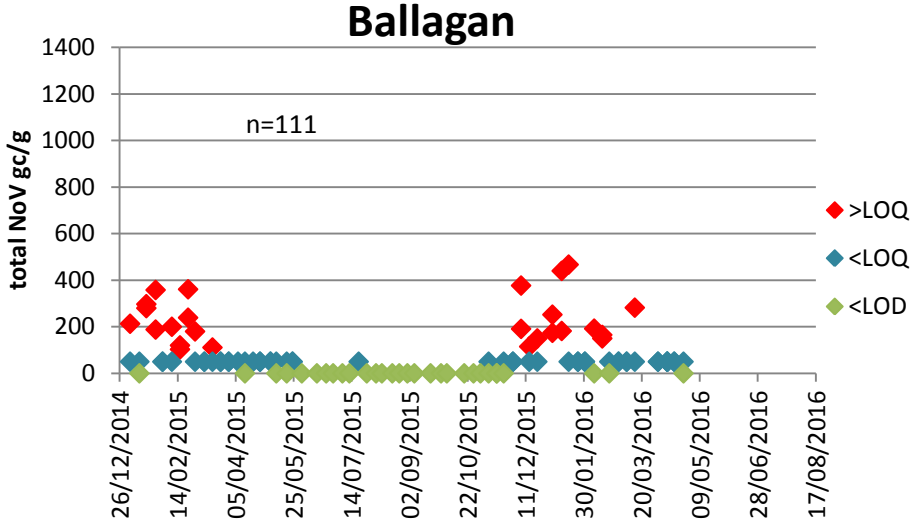
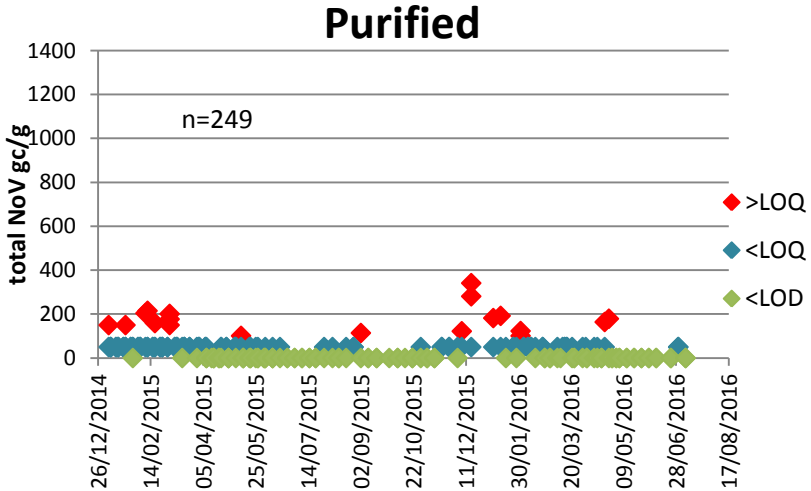
Ongoing monitoring of Carlingford oysters

Jan 2015- July 2016







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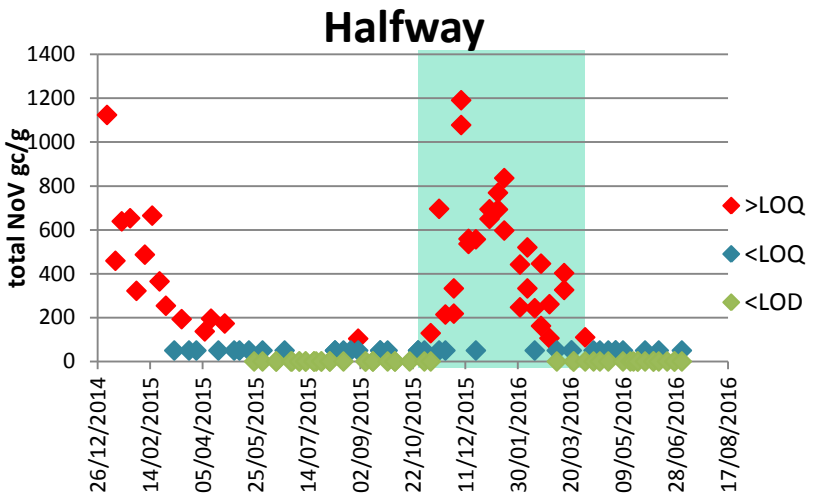
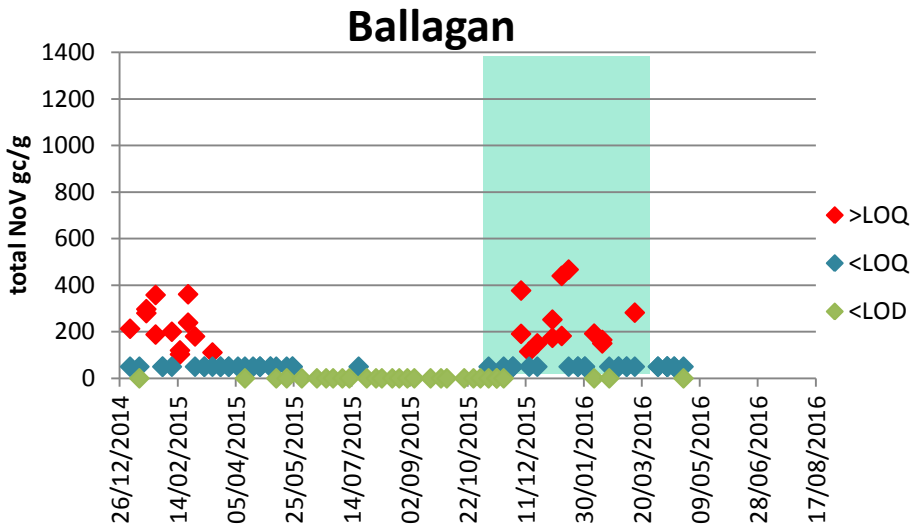
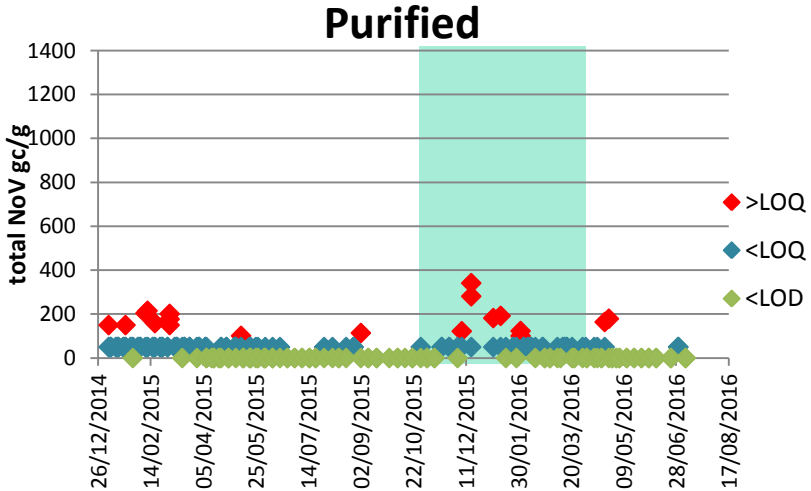
Winter 2015/2016







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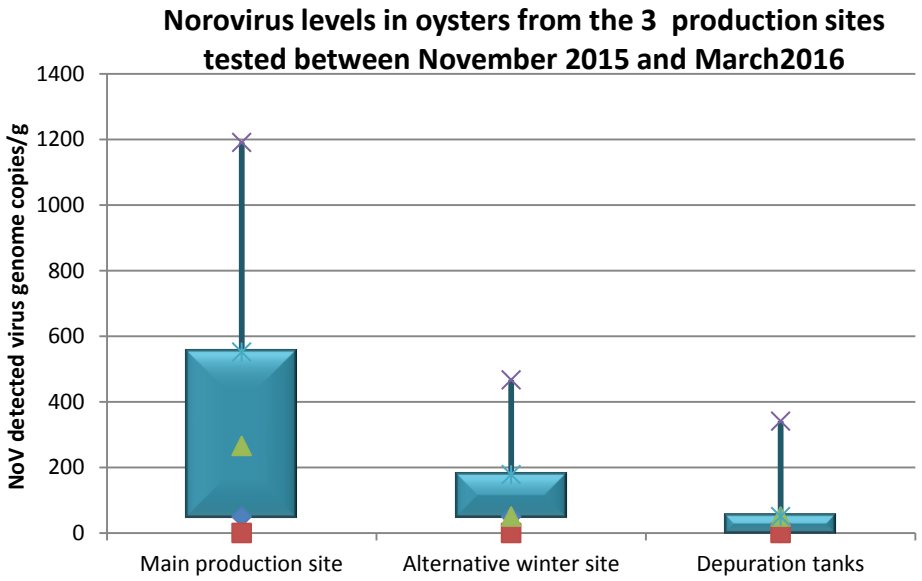
Effect of re-laying



Stats	Main production site	Alternative winter site	Depuration tanks
Q1	50	50	0
Min	0	0	0
Median	266	50	50
Max	1191	467	341
Q3	552	178	50

The use of targeted production procedures (relocation & depuration) significantly reduced the concentration of detectable NoV in oysters and subsequent exposure for consumers.

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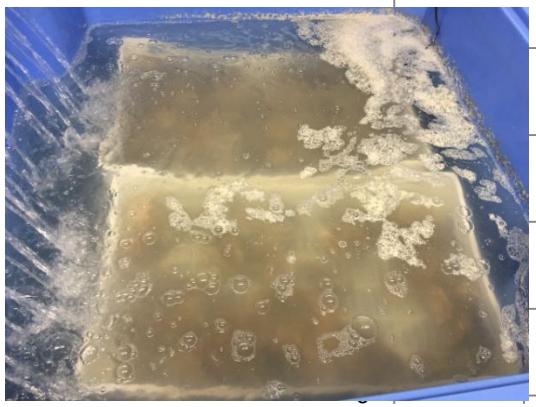
Depuration tanks

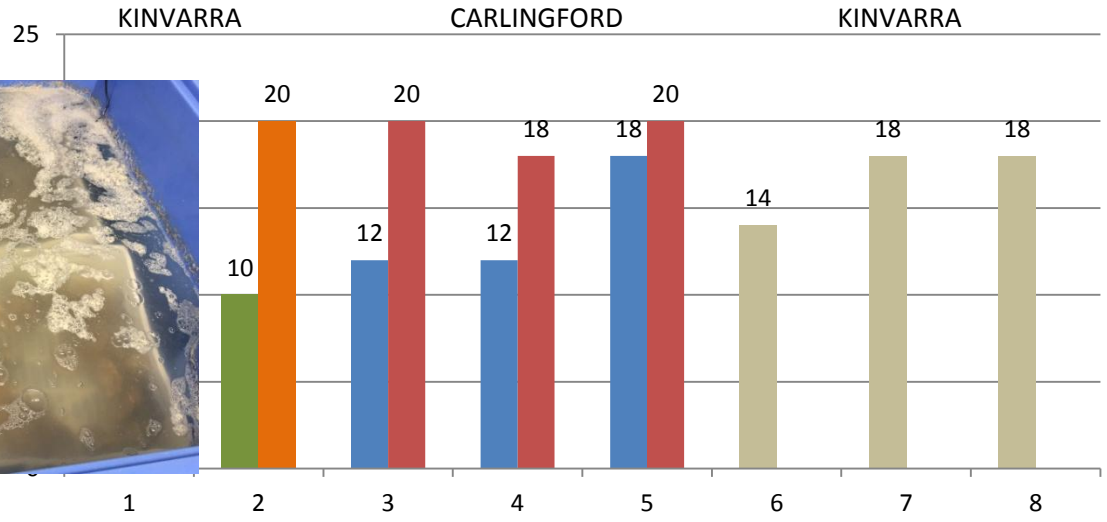
- ✓ ~450L of artificial sea water per tank.
- ✓ Salinity ~28-30ppt .

- ✓ Water temperature controlled with aquarium heaters and external chiller.
- ✓ Room temperature controlled (optional).

Depuration experiments completed to date

Depuration No.	Origin of oysters	Date commenced	Date completed	Tank 1 temp.	Tank 2 temp.	Comments
1	Kinvarra	9.11.2015	16.11.2015	10	12	Spawned, not completed
2	Kinvarra	2.12.2015	9.12.2015	10	20	Spawned, reabsorbed, completed but not sure on relevance of data
3	Carlingford - halfway	11.1.2016	19.1.2016	12	20	-
4	Carlingford - halfway	25.1.2016	1.2.2016	12	18	-
5	Carlingford - halfway	16.2.2016	23.2.2016	18	20	-
6	Kinvarra - 30 individual oysters	11.3.2016	14.3.2016	14	x	-
7	Kinvarra - 30 individual oysters	4.4.2016	8.4.2016	18	x	-
8	Kinvarra - 30 individual oysters	4.4.2016	12.4.2016	18	x	-





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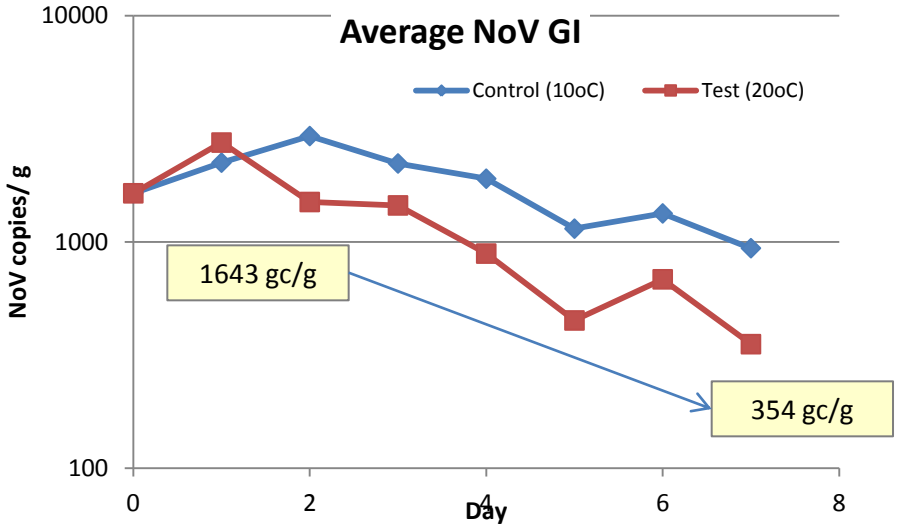
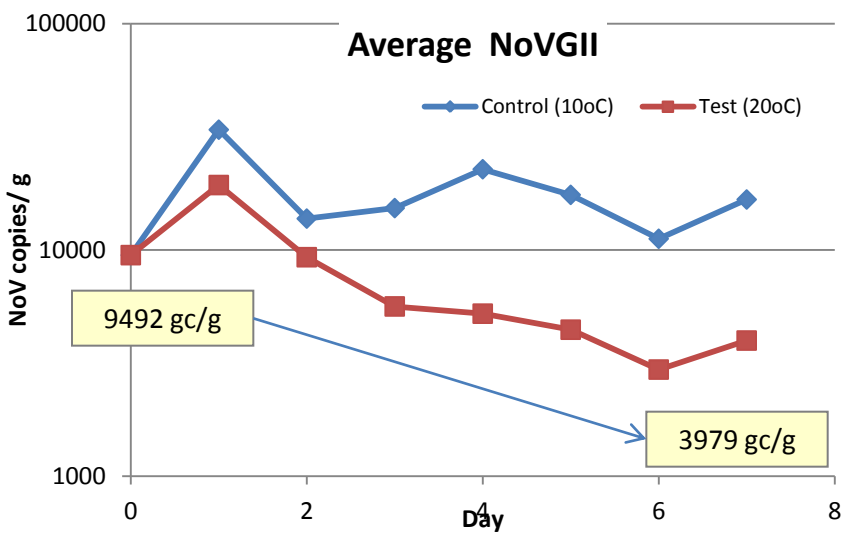
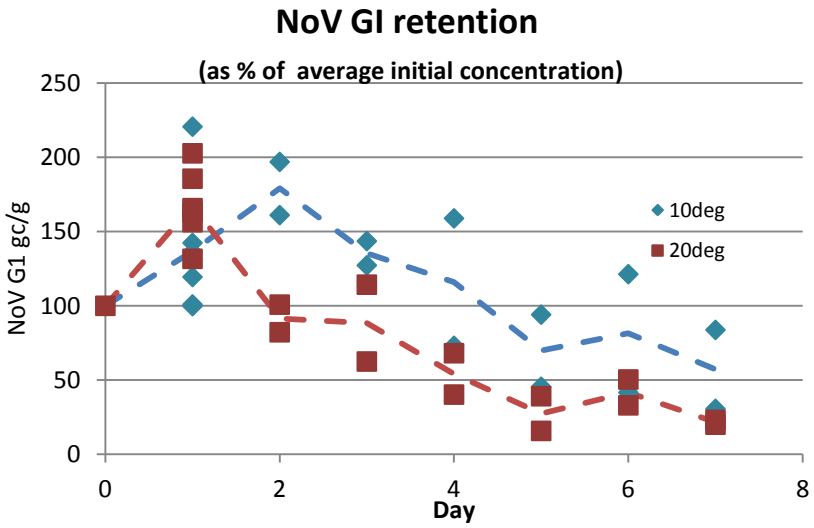
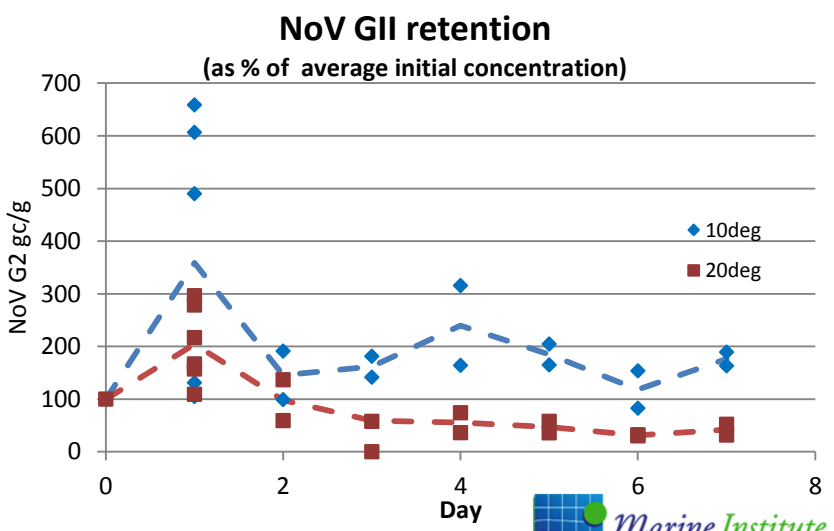
Depuration experiment 2

Average of 2 samples









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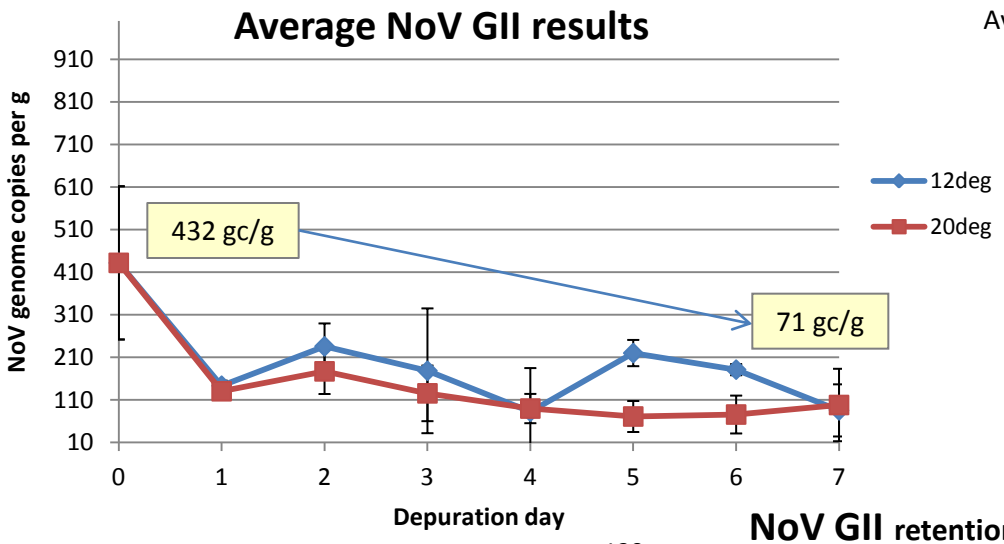
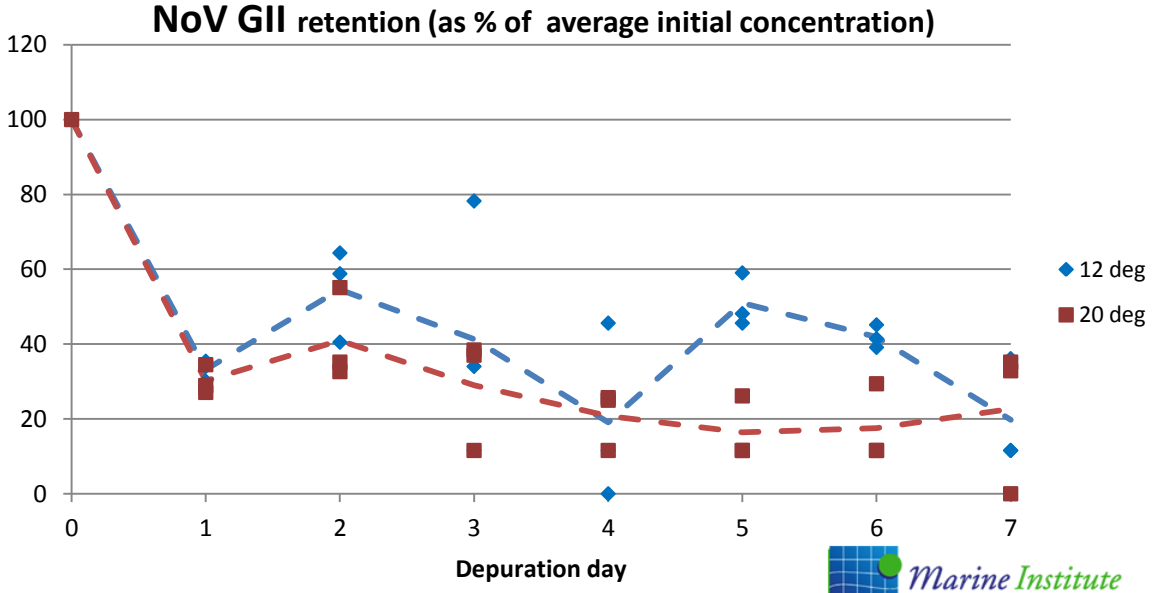
Depuration experiment 3



Average of 3 samples tested



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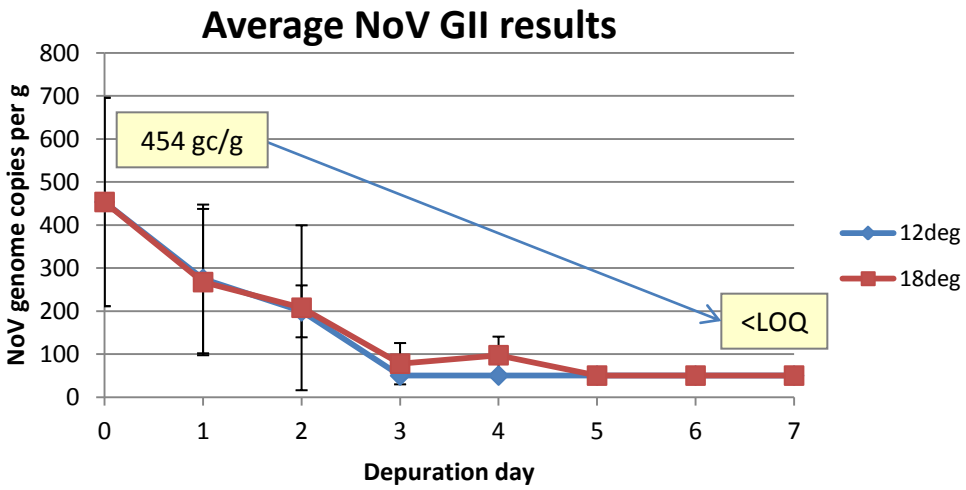
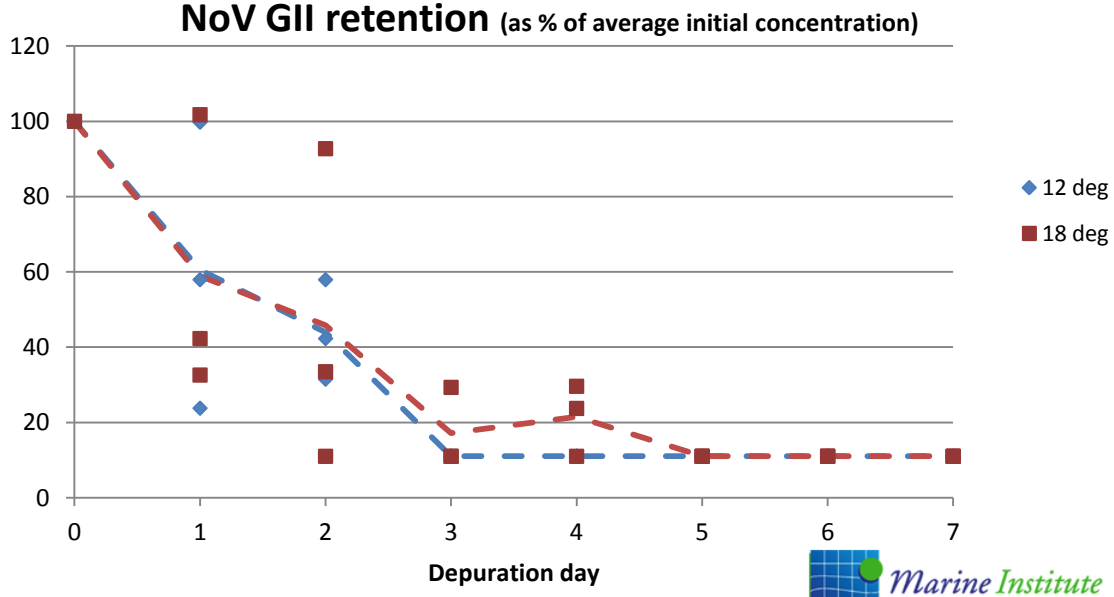
Depuration experiment 4





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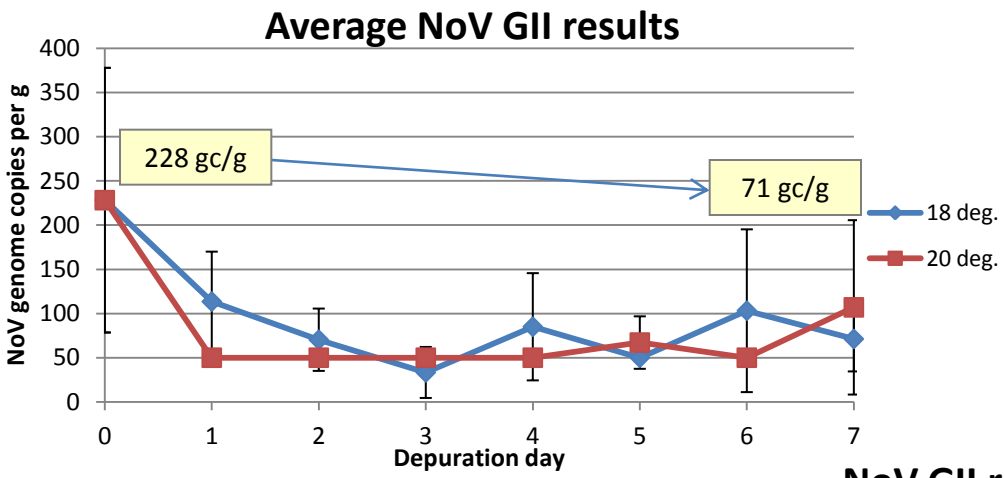
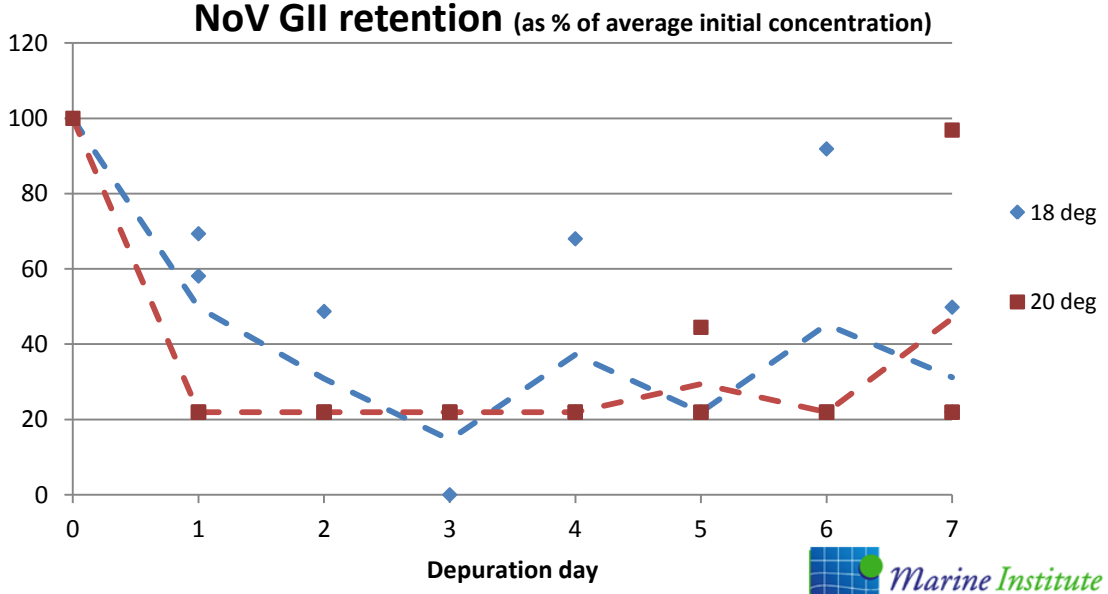
Depuration experiment 5





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Depuration experiments 3 to 5

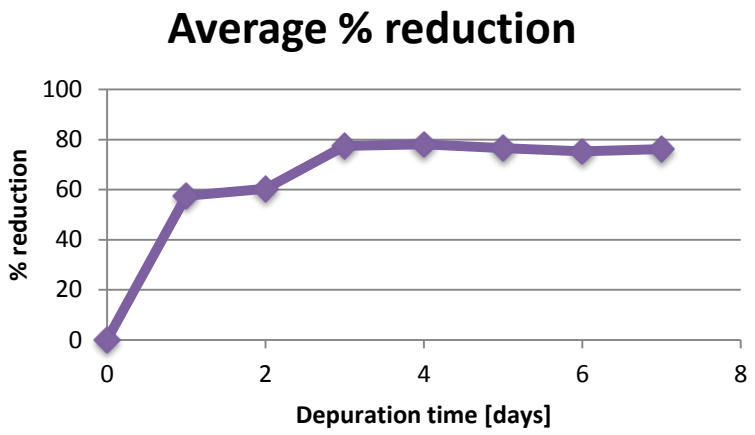
Average NoVGII virus copies/g						
Day	12 deg (1)	20deg (1)	12deg (2)	18deg(1)	18deg(2)	20deg (2)
0	432	432	454	454	228	228
1	143	130	275	267	114	50 (<LOQ)
2	236	177	199	208	70*	50 (<LOQ)
3	178	125	50 (<LOQ)	78*	33*	50 (<LOQ)
4	82*	90*	50 (<LOQ)	97*	85*	50 (<LOQ)
5	220	71*	50 (<LOQ)	50 (<LOQ)	50 (<LOQ)	67*
6	181	76*	50 (<LOQ)	50 (<LOQ)	103	50 (<LOQ)
7	85*	98*	50 (<LOQ)	50 (<LOQ)	71*	107

NoVGII % retention							Av. % reduction
Day	12 deg (1)	20deg (1)	12deg (2)	18deg(1)	18deg(2)	20deg (2)	
0	100	100	100	100	100	100	0
1	33	30	61	59	50	22	58
2	55	41	44	46	31	22	60
3	41	29	11	17	15	22	77
4	19	21	11	21	37	22	78
5	51	16	11	11	22	29	77
6	42	18	11	11	45	22	75
7	20	23	11	11	31	47	76

Results show an average of 3 samples tested per each time point.

All <LOQ results were assigned a value of 50 copies/g.

* One or two out of 3 samples tested as <LOQ.



- Depuration reduces the concentration of NoV in oysters but does not remove the virus completely.
- At this stage it's difficult to tell how significant temperature is.
- Need more contaminated oysters to stay above the LOQ levels.

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Future work

- Repeat depuration experiments with oysters contaminated with higher levels of NoV.
  - Move a batch of Carlingford oysters to Kinvarra?
  - Use other source of oysters (avoid diploids as they will likely spawn at elevated water temperatures)
- Estimate the infectivity of NoV in samples pre- and post- depuration using FRNA bacteriophage as a surrogate.

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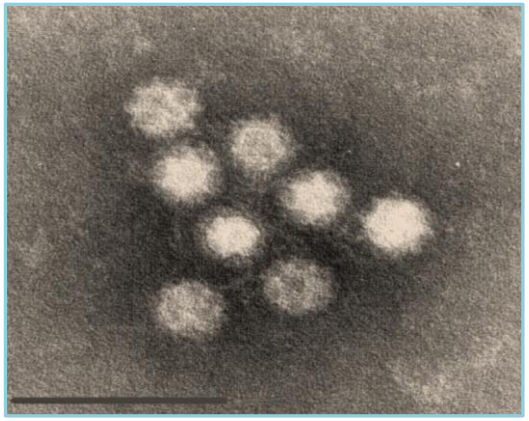
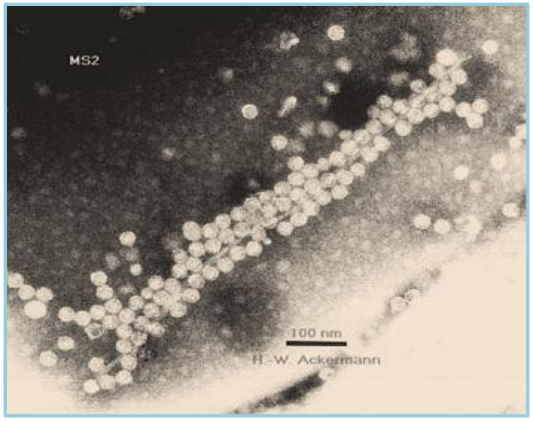
Why bacteriophage?





Norovirus (*Caliciviridae*)

- ssRNA
- Simple cubic capsid
- 25-30 nm
- Found in a wide range of mammalian species
- Real-time PCR assay only, **no infectivity assays available**

FRNA Phage (*Leviviridae*)

- ssRNA
- Simple cubic capsid
- 25-30 nm
- Found in a wide range of mammalian species
- Real-time PCR assay and **viability/infectivity assay available**

John Flannery, 2011

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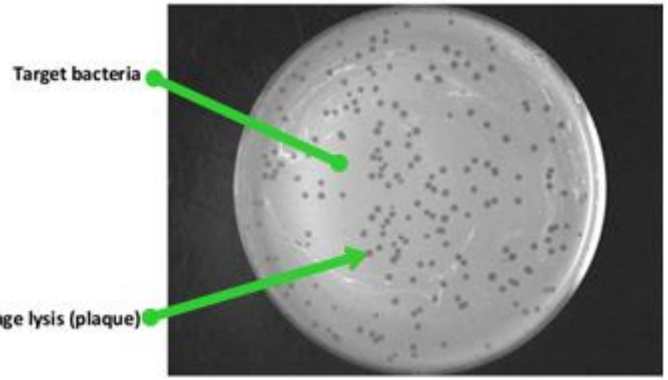
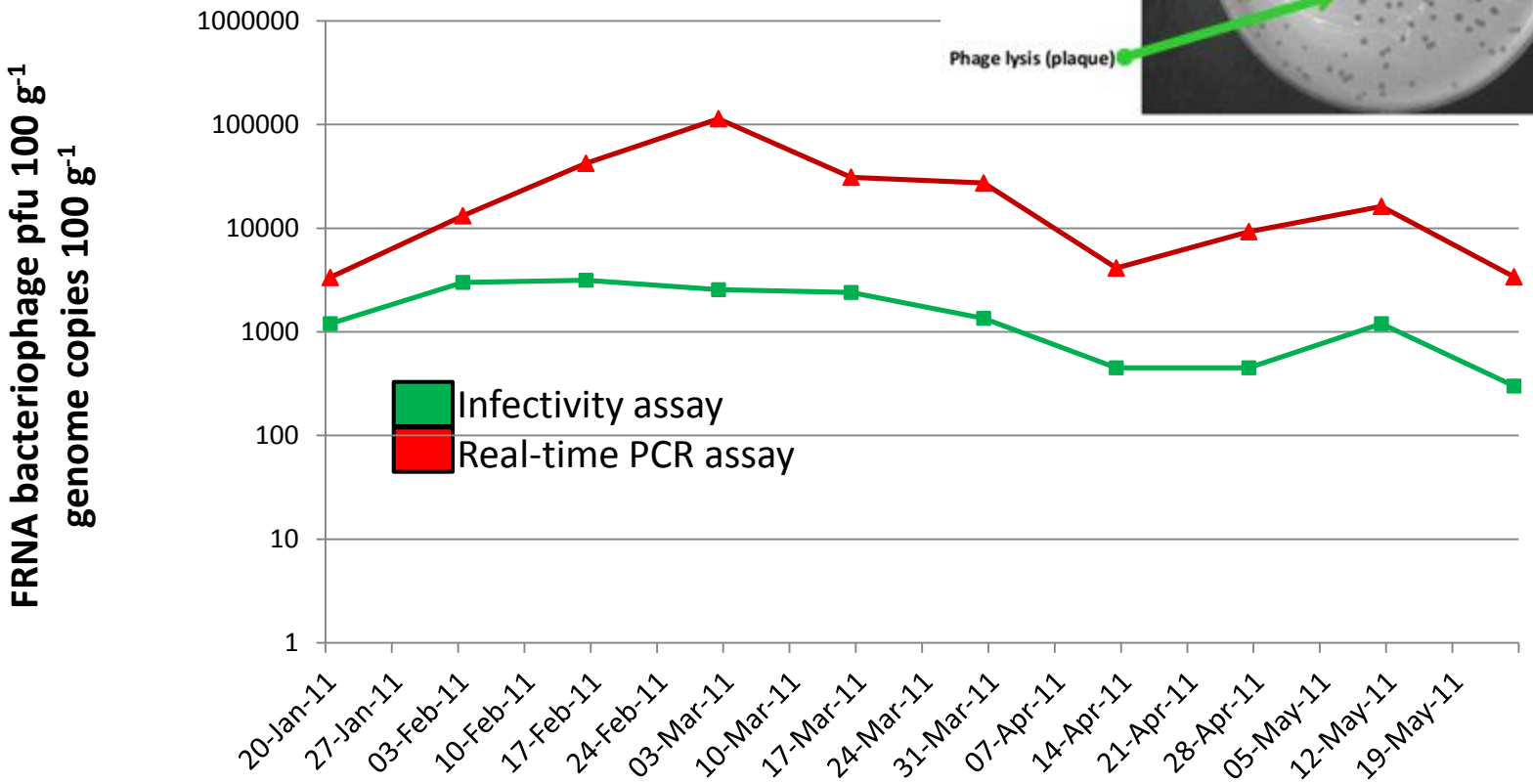
Methods of detection





John Flannery, 2011

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