

# GENOMIC SCIENCE TOOLS BEING IMPLEMENTED ON SAMPLES FROM THE FIRST GULF OF ALASKA EXPEDITION IN 2019

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PICES - W16



# Wild salmon declines – what is killing fish in the ocean?



## Is a fish is stressed or unhealthy?

- Mortalities not observable in the ocean
- Behavioral shifts/clinical signs go unnoticed
- Sampling restricted to “live” fish -> gear selectivity
- Compromised fish have higher risk of predation

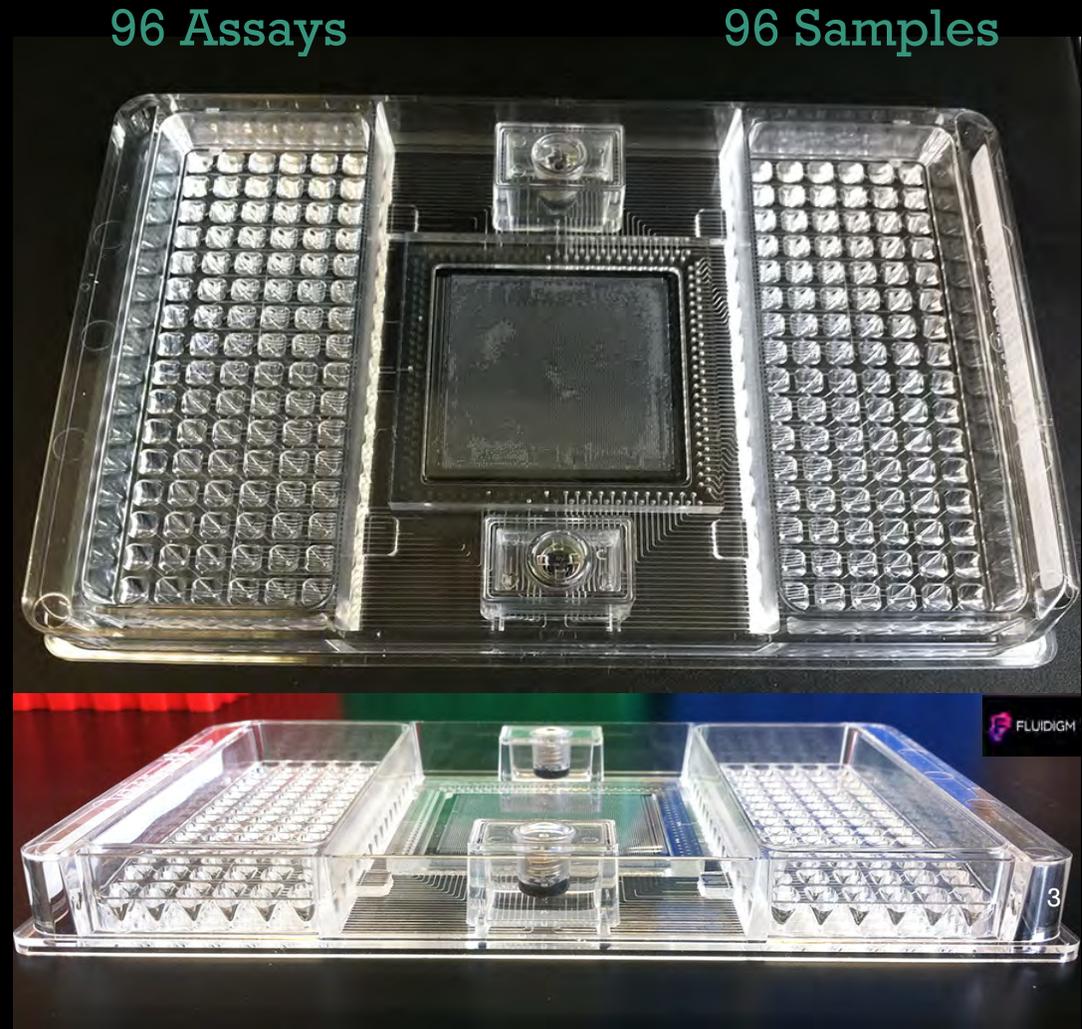
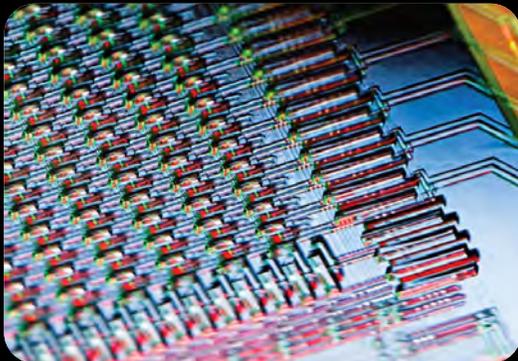
## Cumulative stressors

- Effect of temperature, salinity, oxygen saturation, etc...
  - Impact on fish?

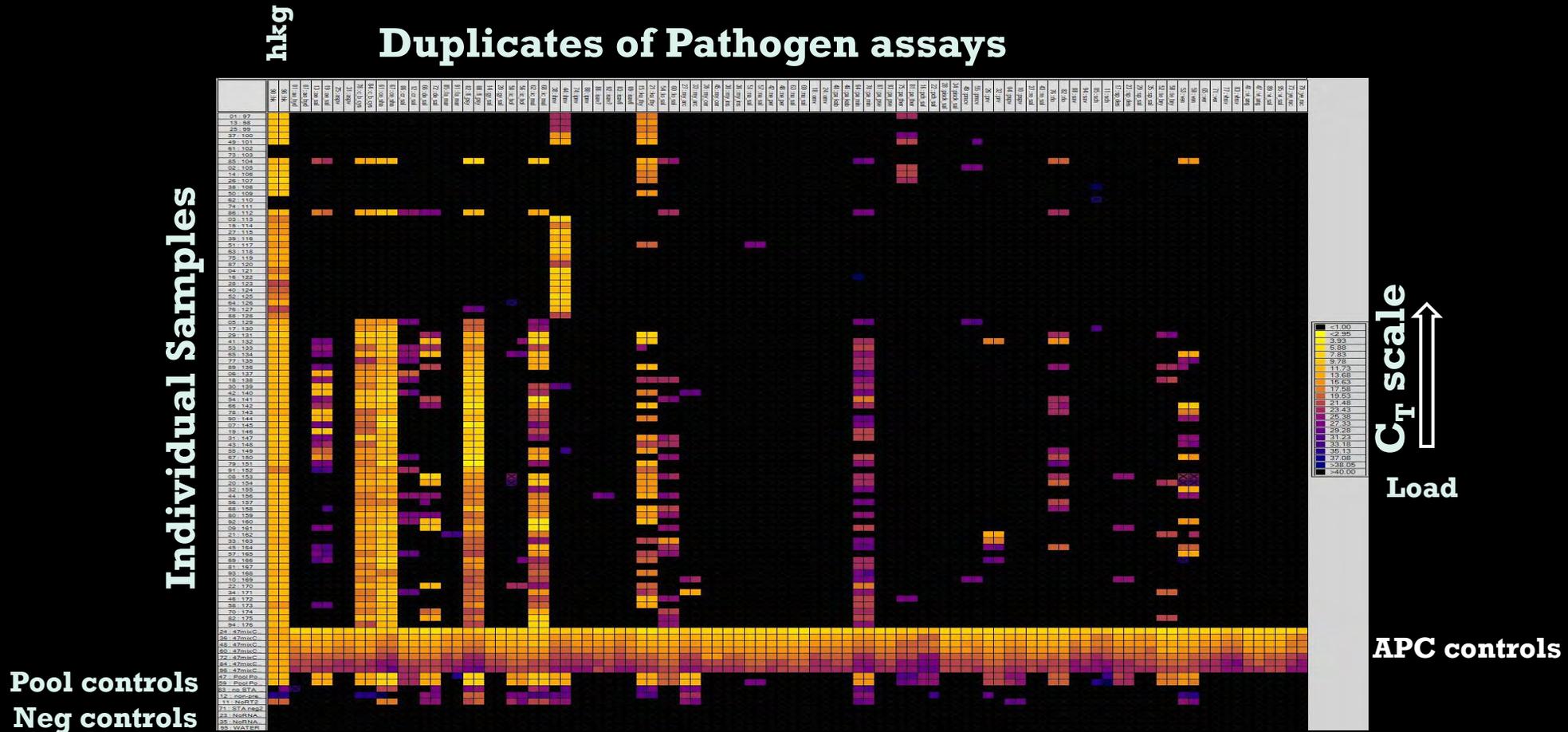
Identify molecular biomarkers that can recognize specific stressor and disease states

# Genomic Tool to address salmon health

- Fluidigm BioMark™ Microfluidics System
  - High Throughput qRT-PCR
  - 96x96 Dynamic Array = 9,216 reactions / run
  - 96 individuals tested against 96 assays
    - Pathogens/harmful algal bloom species
    - Host gene expression



# Fluidigm Biomark: Pathogen detection

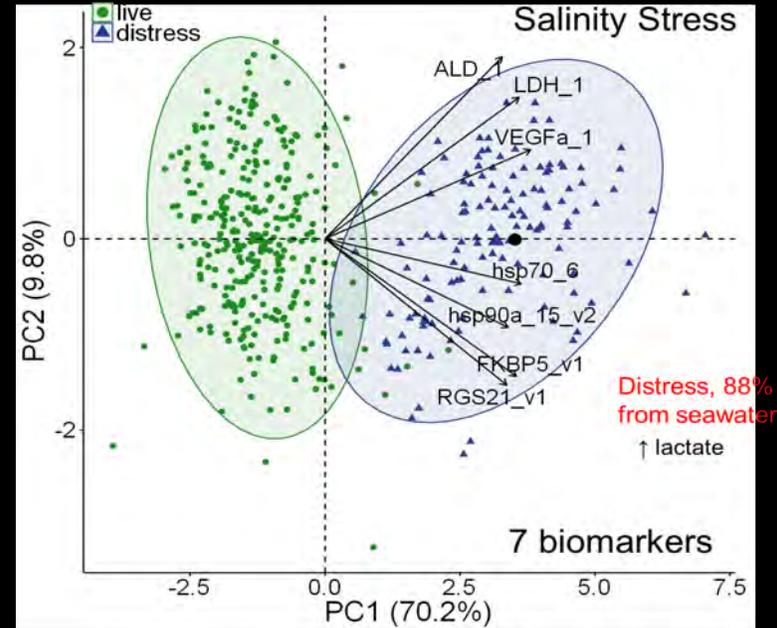
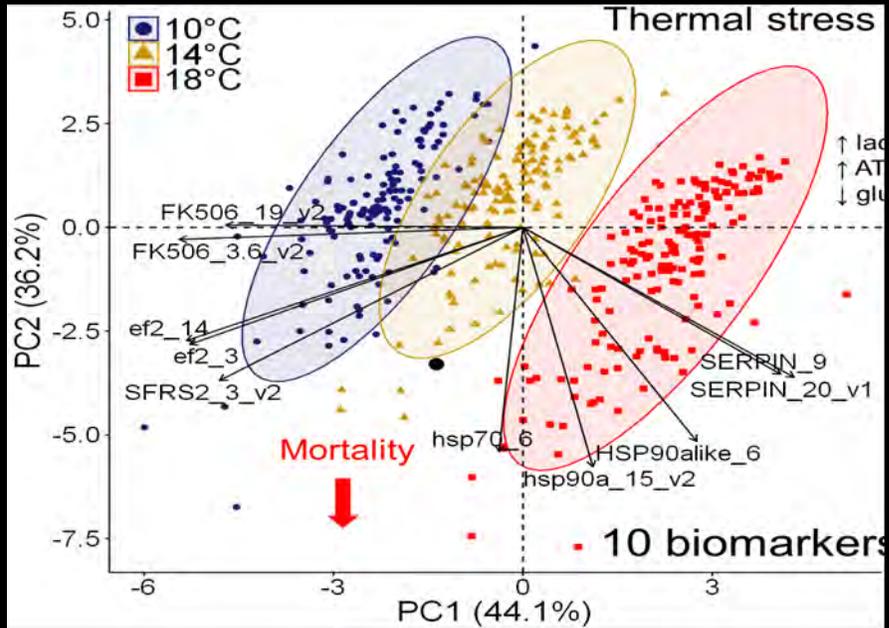




# Health and Condition Salmon Fit-Chips

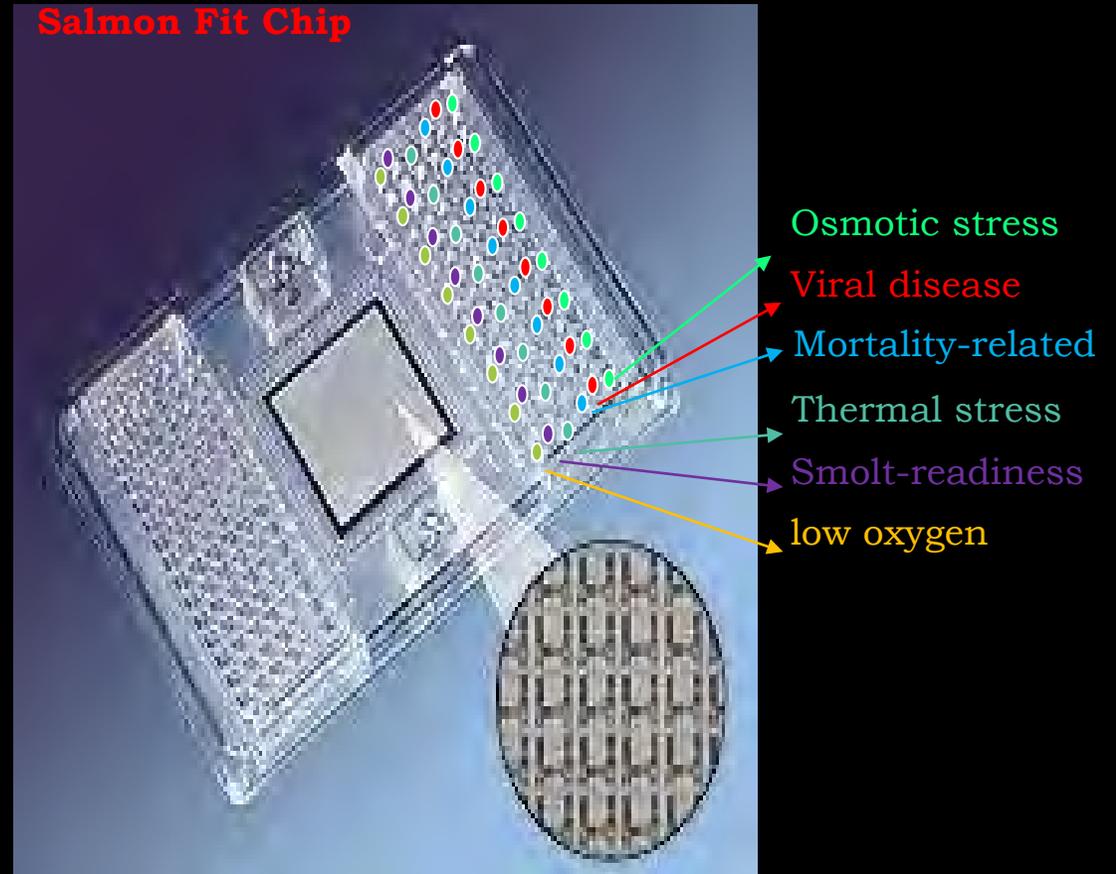


- Non-destructive gill clips to characterize stress and infection
- Biomarker panels of 8-10 genes per stressors/disease state
  - From transcriptome challenge studies
  - Validated in laboratory trials



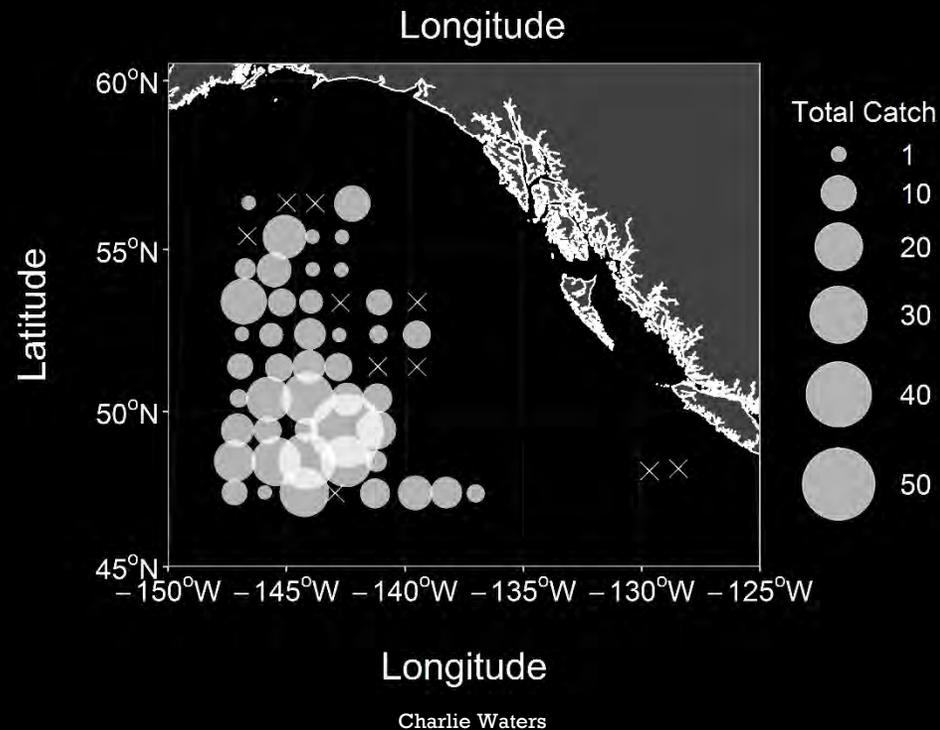
# Fluidigm Biomark: Stressor Detection

- Curated host biomarker panels to identify specific stressor/disease states
  - Thermal stress
  - Osmotic stress
  - Hypoxia stress
  - Inflammation
  - Viral disease
  - Immune status
  - Imminent Mortality



# IYS GoA Expedition – Unique insights into the salmon lifecycle

- Poorly understood oceanic phase of salmon lifecycle
- May contribute to variable ocean returns
  - As critically as early coastal marine life?
- Compare “health” to coastal margin



# IYS GoA - Sample Collection

- Collected and dissected 255 salmon
  - Coho 80/94
  - Chum 84/223
  - Chinook 3/3
  - Pink 27/31
  - Sockeye 61/73
- Assistance:
  - **Alexey, Albina, Svetlana**



# Pathogen detection in overwintering GoA Salmon

- Tested for 47 pathogens
  - 20 viruses (\*many newly characterized in BC salmon)
  - 16 parasites
  - 11 bacteria

*Aeromonas salmonicida*

*Candidatus Branchiomonas cysticola*

*Piscichlamydia salmonis*

*Piscirickettsia salmonis*

*Renibacterium salmoninarum*

Rickettsia-like organism (RLO)

Gill chlamydia (Sch)

*Tenacibaculum maritimum*

*Vibrio anguillarum*

*Vibrio salmonicida*

*Yersinia ruckeri*

*Ceratanova shasta*

*Dermocystidium salmonis*

*Ichthyophonus hoferi*

*Ichthyophthirius multifiliis*

*Kudoa thyrsites*

*Loma salmonae*

*Myxobolus arcticus*

*Myxobolus insidiosus*

*Nanophyetus salmincola*

*Neoparamoeba perurans*

*Parvicapsula kabatai*

*Parvicapsula minibicornis*

*Parvicapsula pseudobranchicola*

*Paranucleospora theridion*

*Sphaerothecum destruens*

*Tetracapsuloides bryosalmonae*

\**Arenavirus I MGL*

\**Arenavirus II MGL*

\**Bafini virus*

\**Circo virus*

\**Coronavirus MGL*

\**Cutthroat Trout Virus MGL*

\**Hantavirus*

\**Nidovirus 2*

\**Orthomyxovirus MGL*

\**Picorna virus*

*Piscine orthoreovirus*

\**Pacific salmon parvovirus*

\**Qin virus*

\**Reovirus MGL*

\**Rhabdo virus*

*SalmovirusWFRC1\_virus*

\**MGL Small unknown RNA Virus*

*Viral erythrocytic necrosis virus*

*Viral encephalopathy and retinopathy virus*

*Viral hemorrhagic septicemia virus*

*Housekeeping gene*

# Prevalence of 20 agents detected in GoA

**Candidatus Branchiomonas cysticola**

**Ceratanova shasta**

**Ichthyophonus hoferi**

**Ichthyophthirius multifiliis**

**Kudoa thyrsites**

**Loma salmonae**

**Myxobolus insidiosus**

**Nanophyetus salmincola**

**Parvicapsula kabatai**

**Parvicapsula minibicornis**

**Parvicapsula pseudobranchicola**

**Paranucleospora theridion**

**Picornavirus\***

**Piscine orthoreovirus**

**Pacific salmon parvovirus\***

**\*Rhabdovirus**

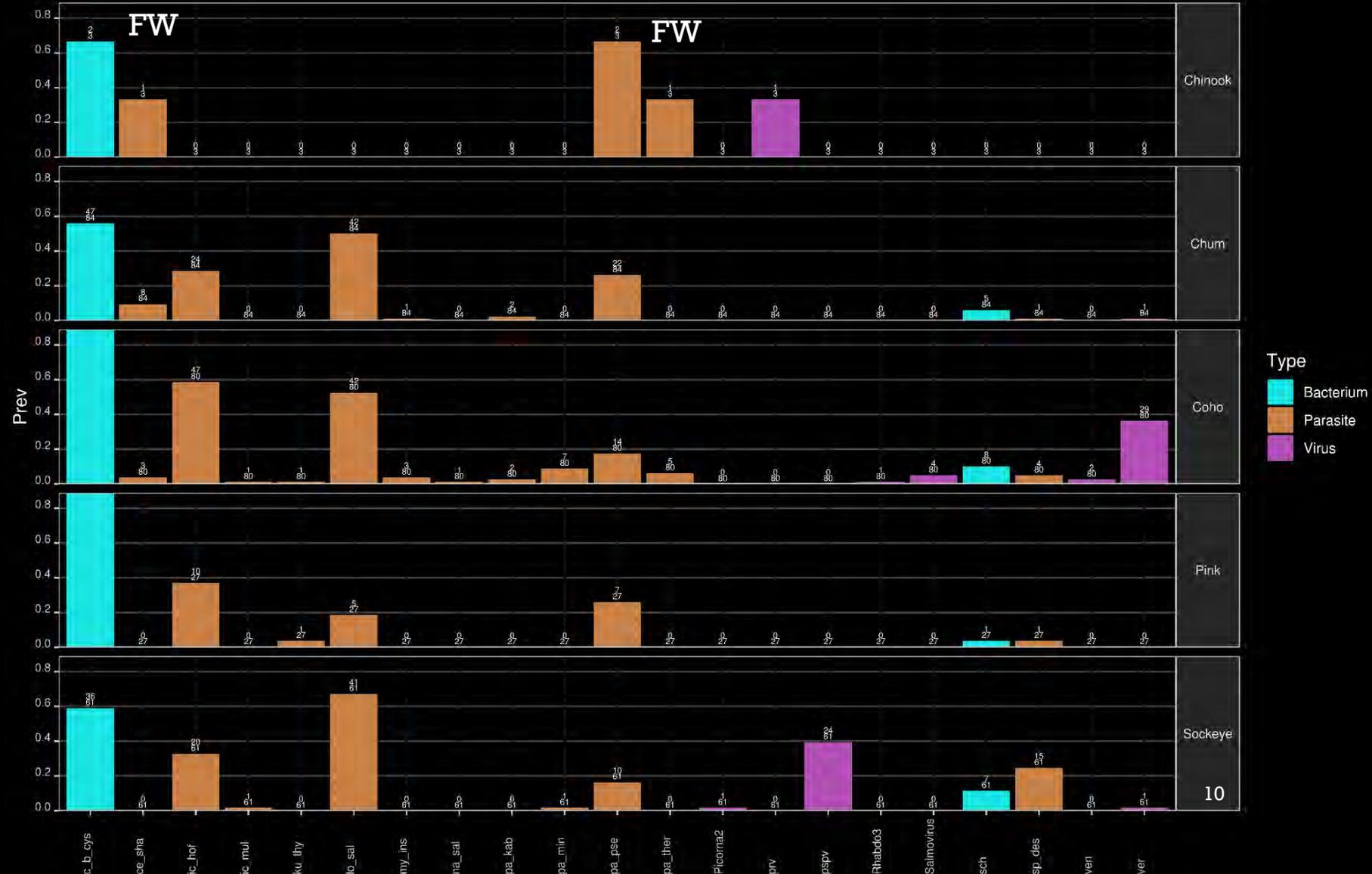
**SalmovirusWFRC1\_virus\***

**Gill chlamydia (Sch)**

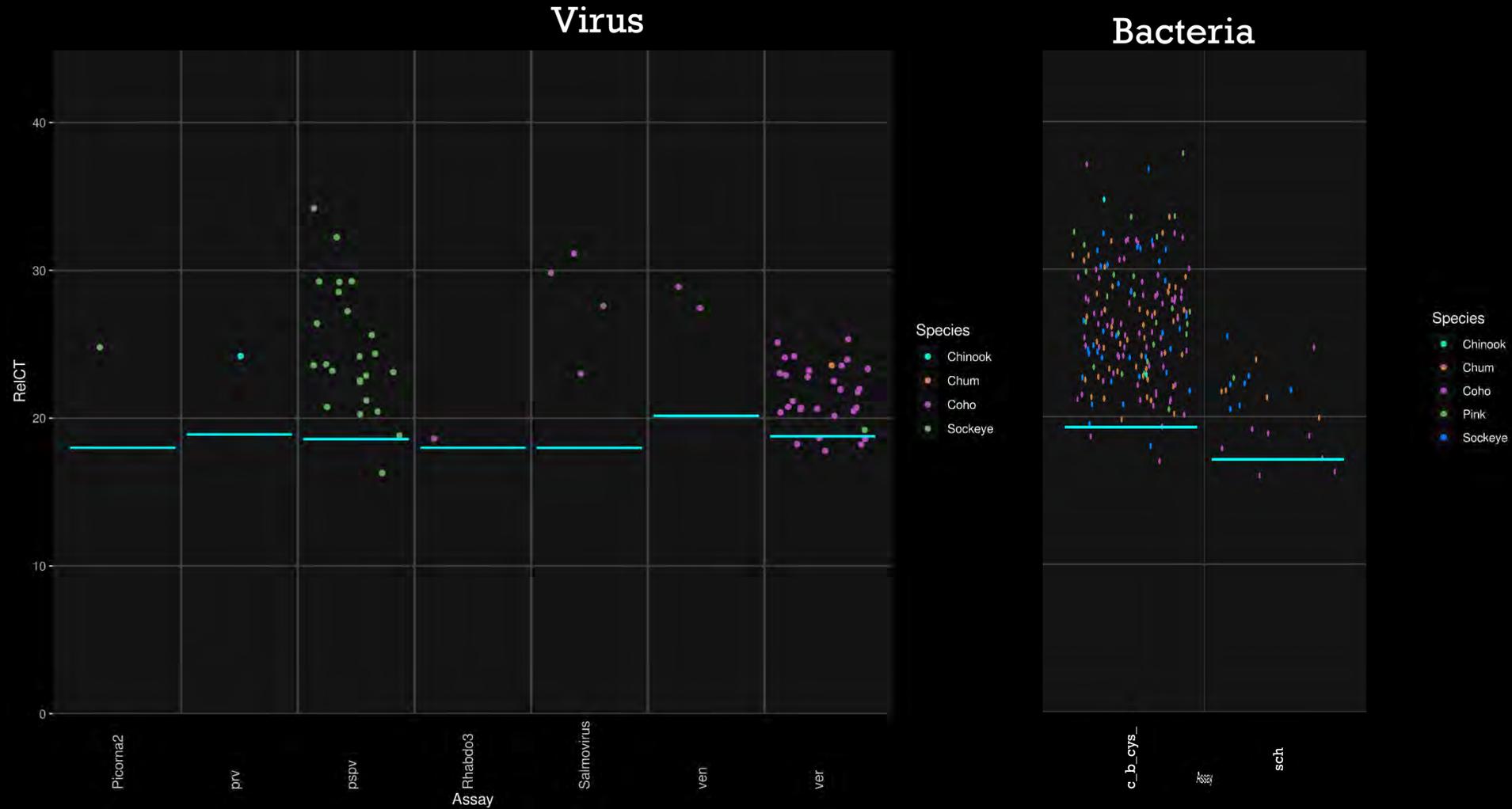
**Sphaerothecum destruens**

**Erythrocytic necrosis virus**

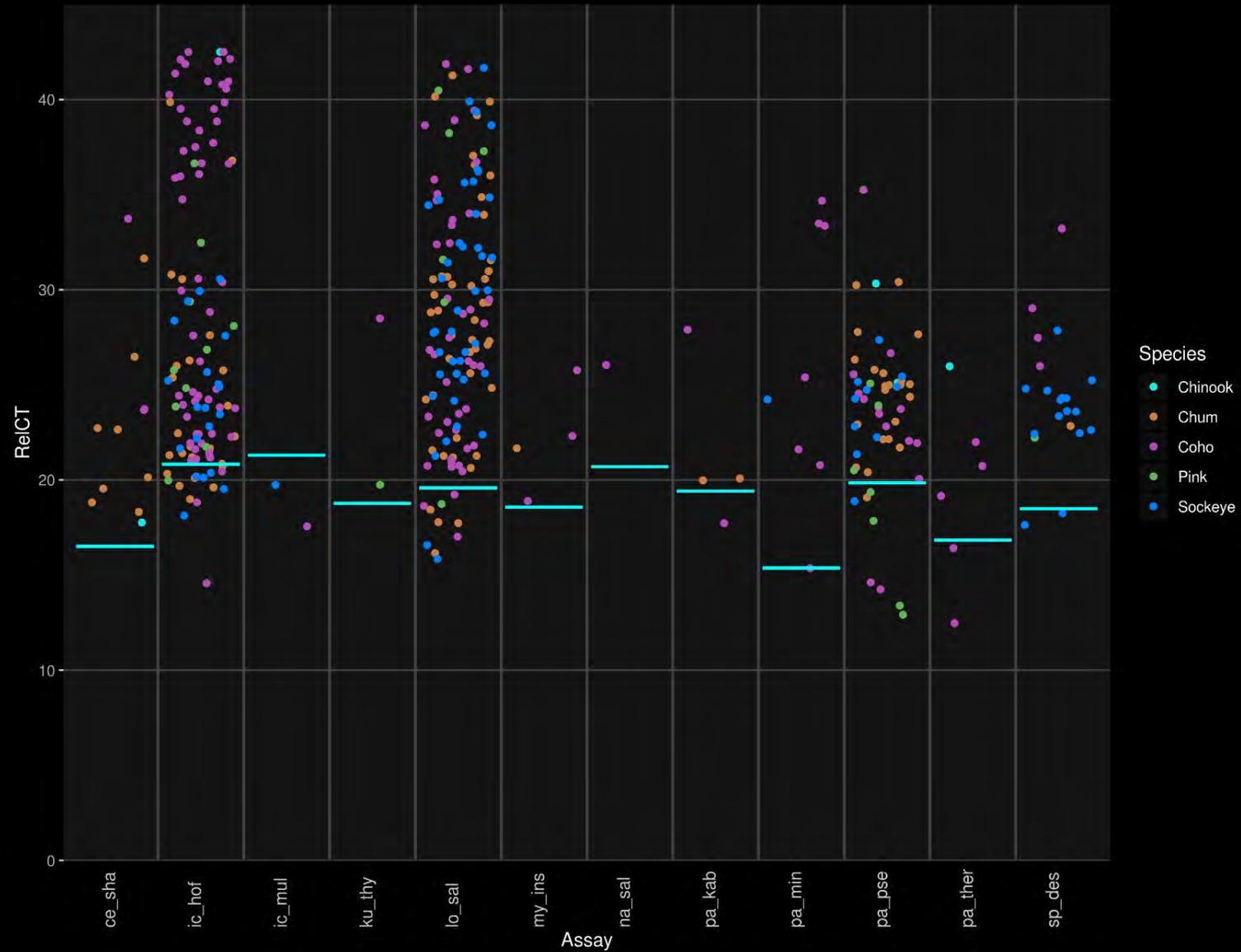
**Encephalopathy and retinopathy virus**



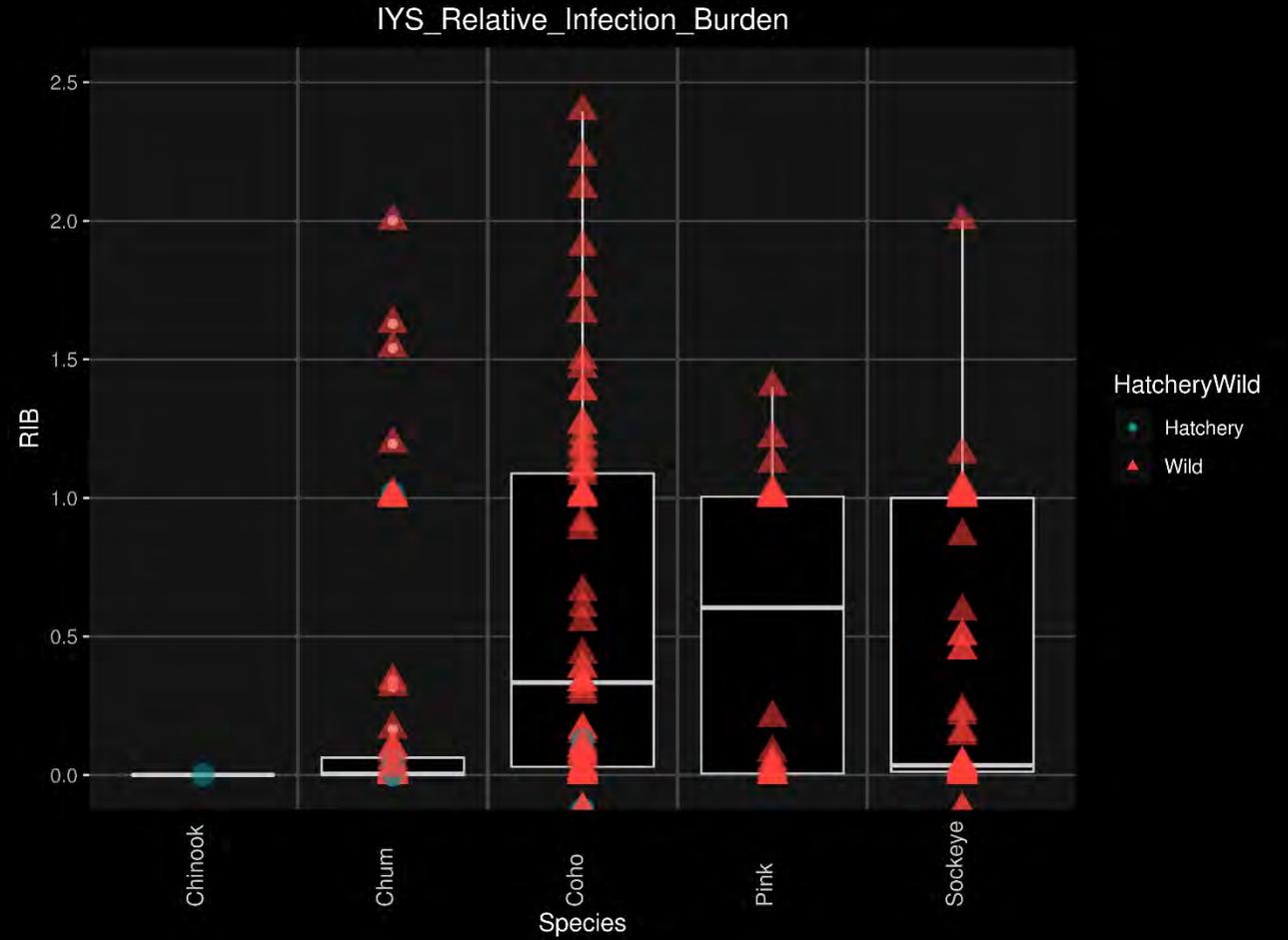
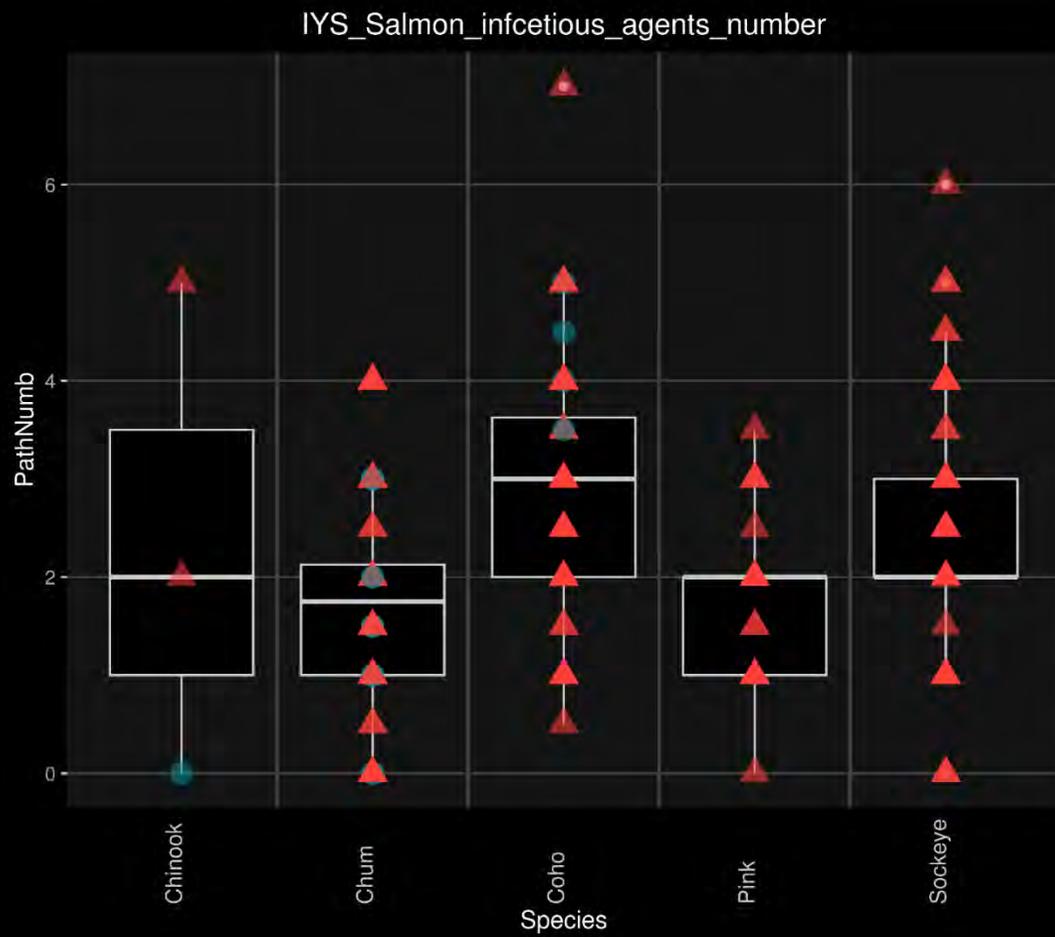
# Pathogen-Load: Viruses - Bacteria



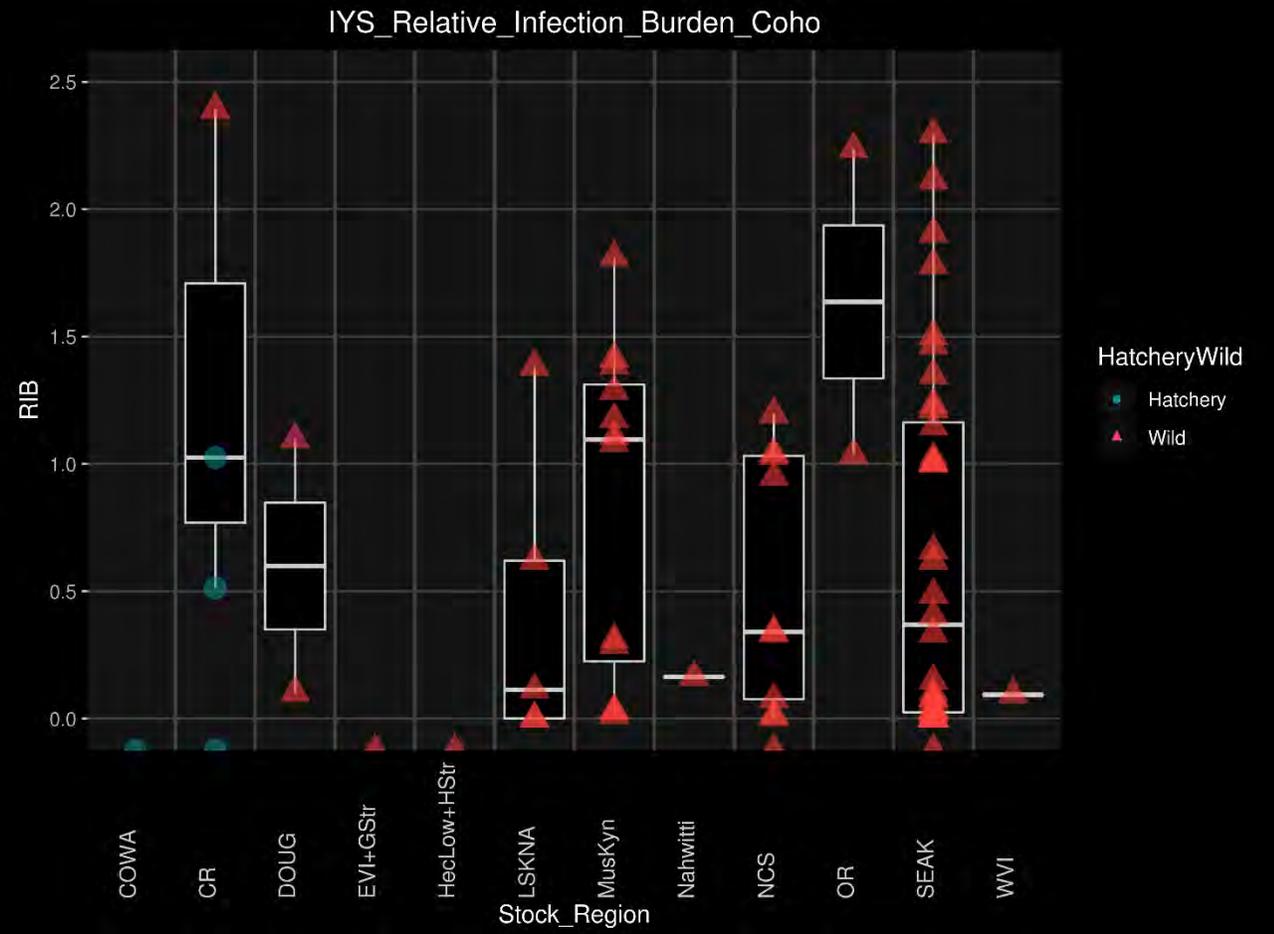
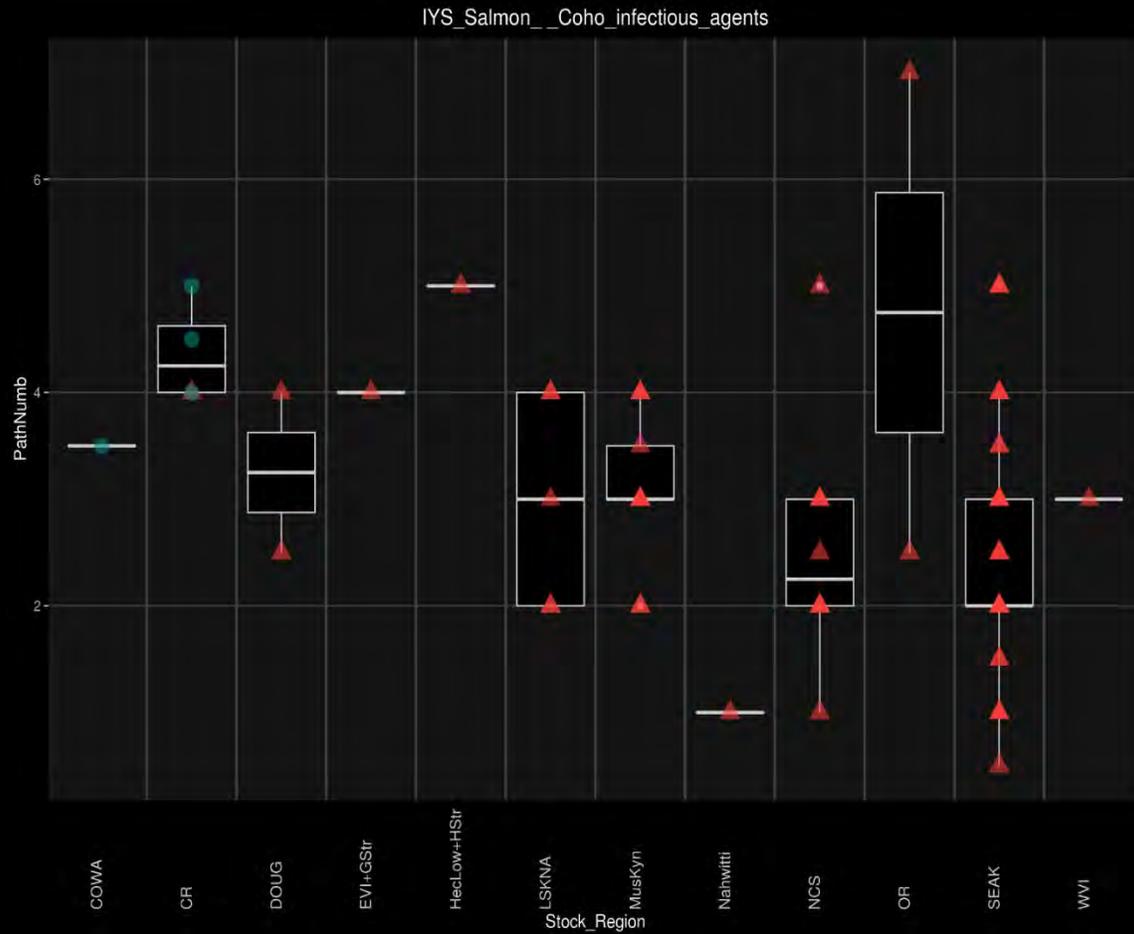
# Pathogen-Load: Parasites



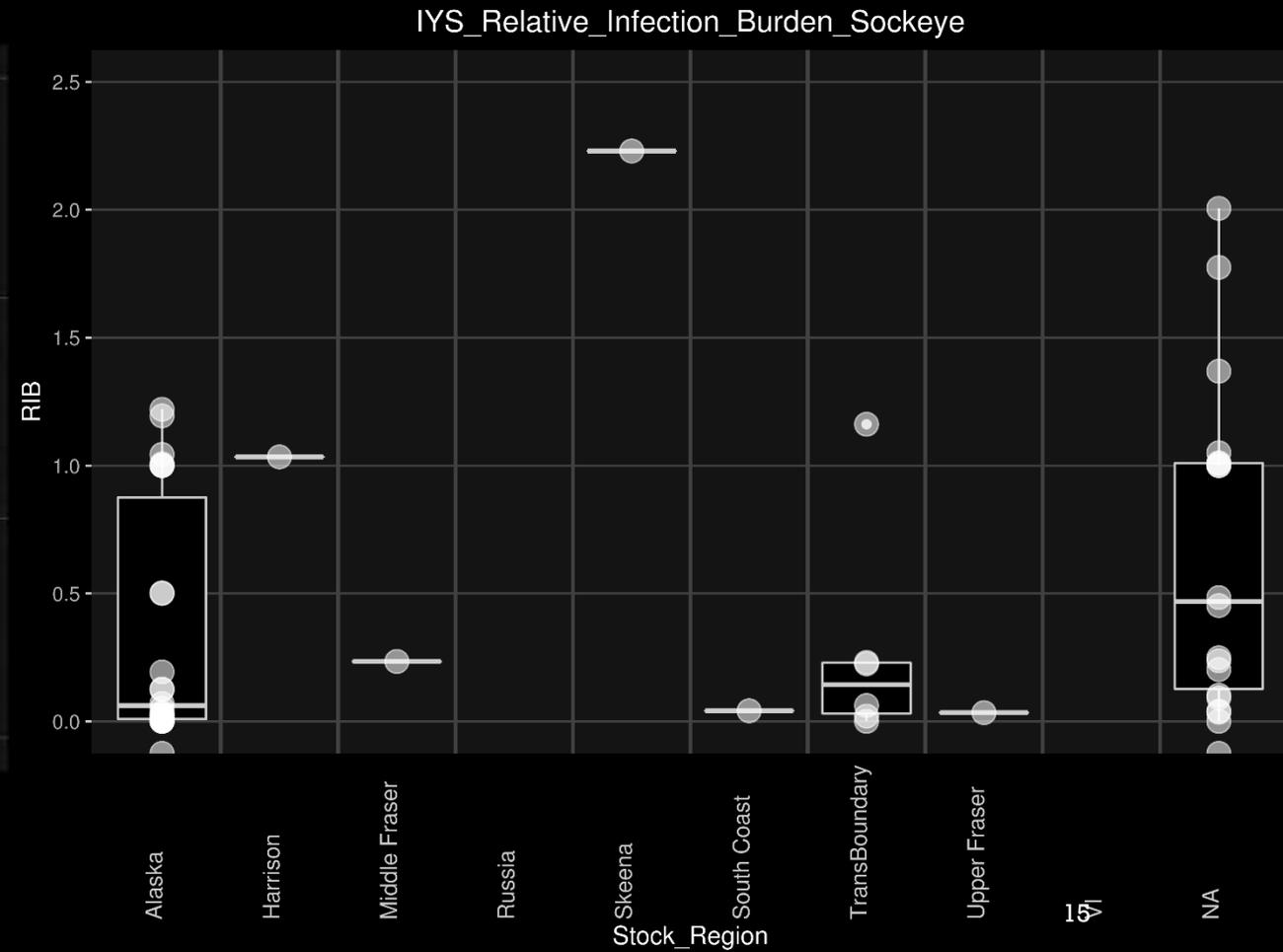
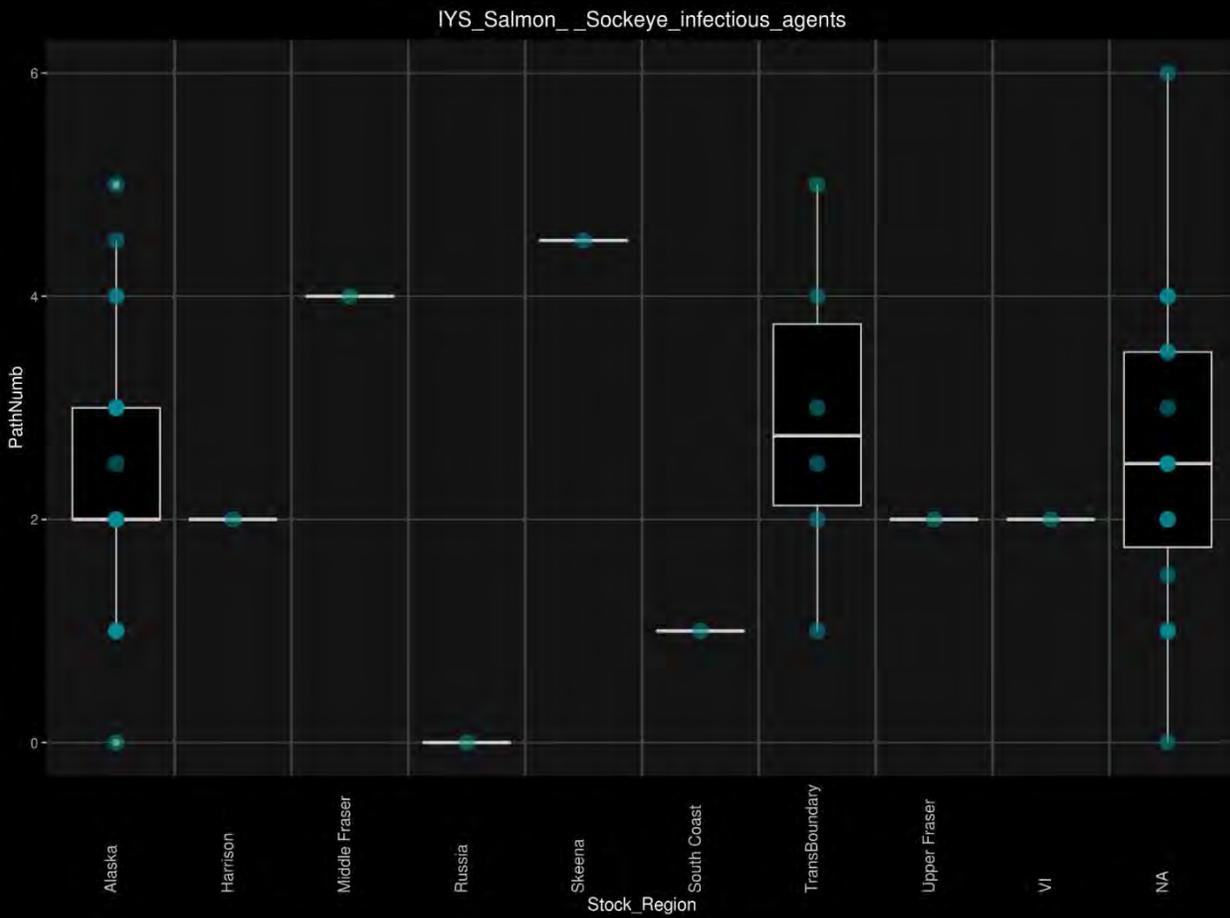
# Pathogen number by species



# Coho pathogens by stock (N=80)



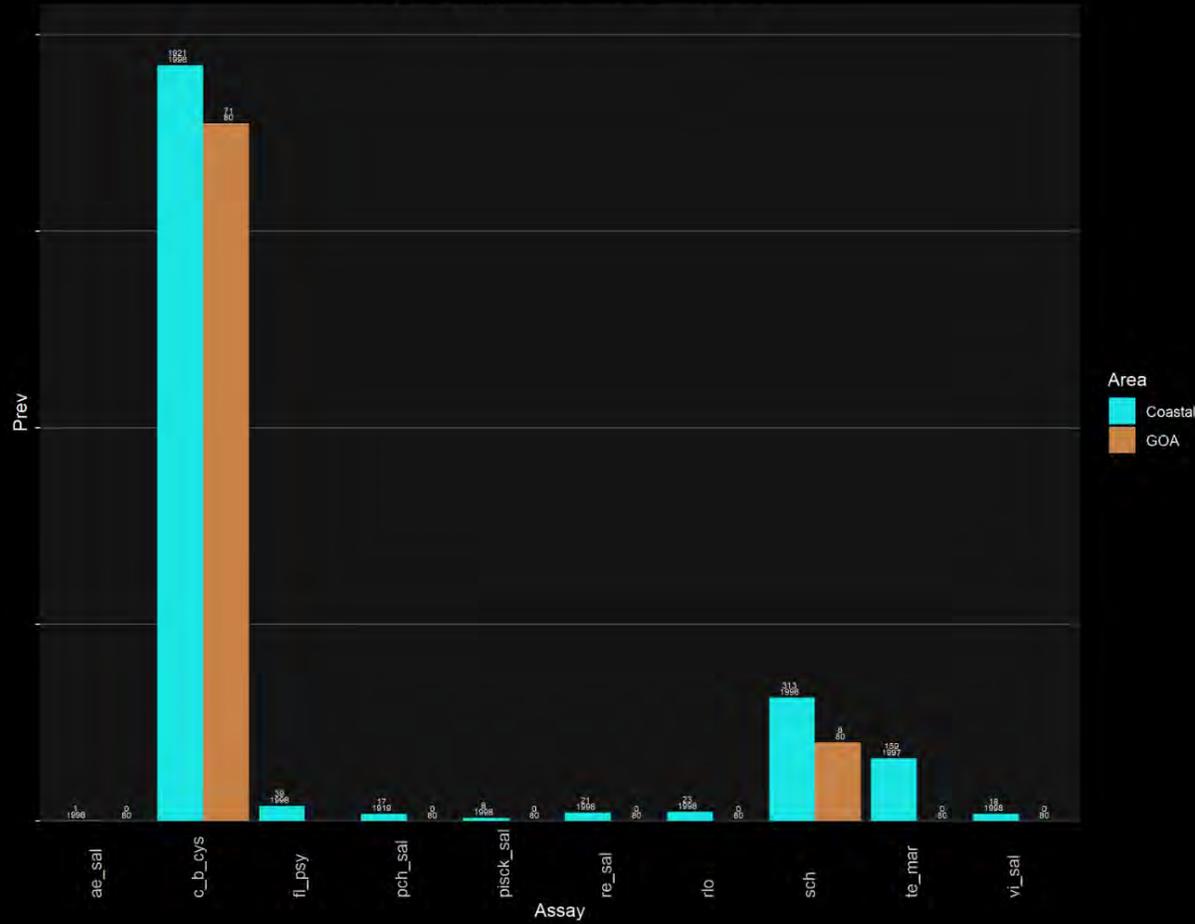
# Sockeye pathogens by stock (N=61)



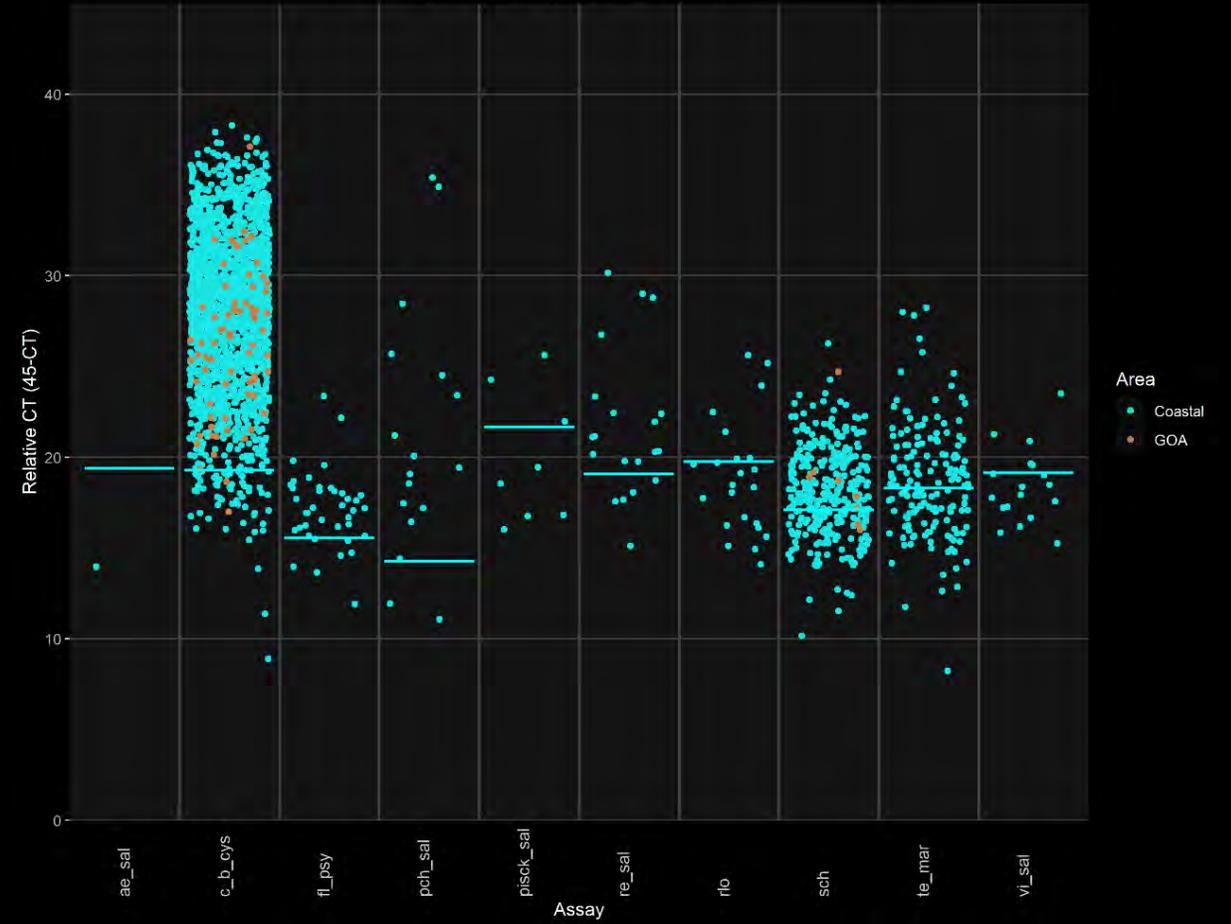


# GoA vs BC Coast: Coho - Bacteria

IYS Coho Comparisons Bacterium No LOD

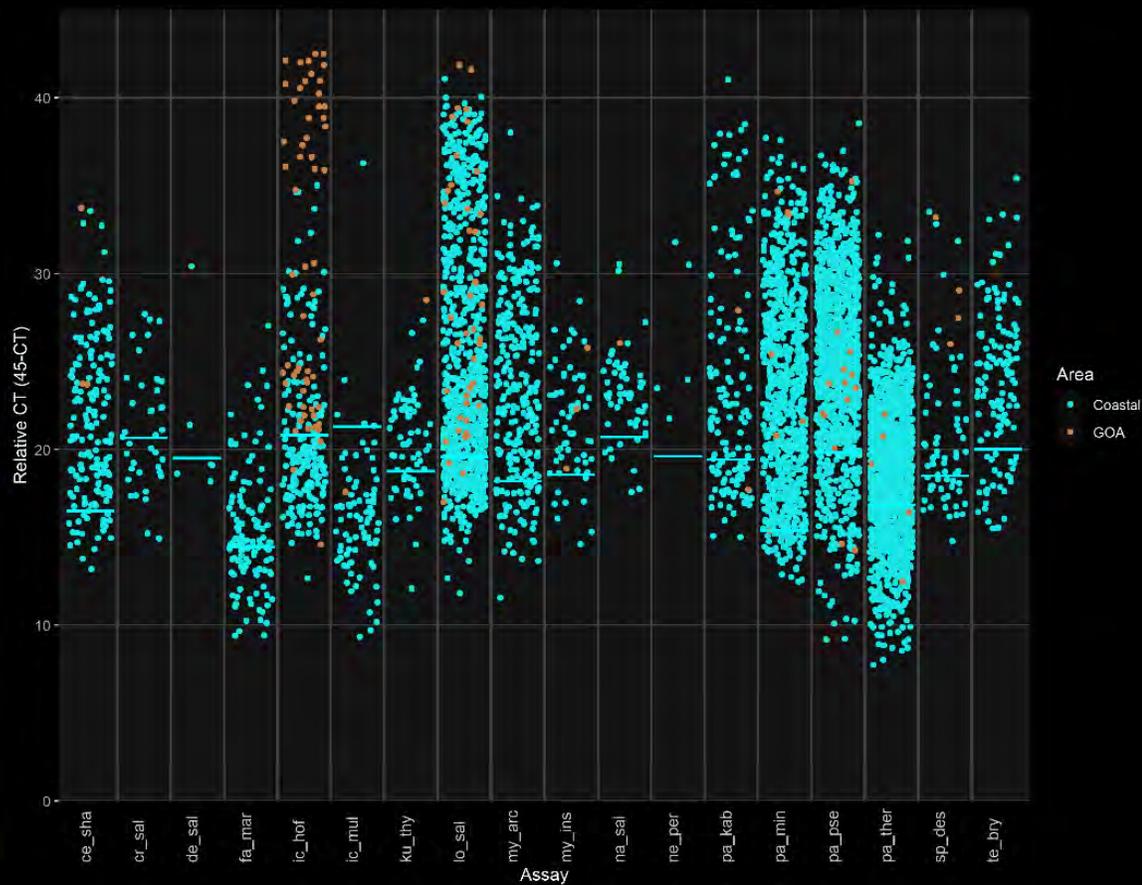
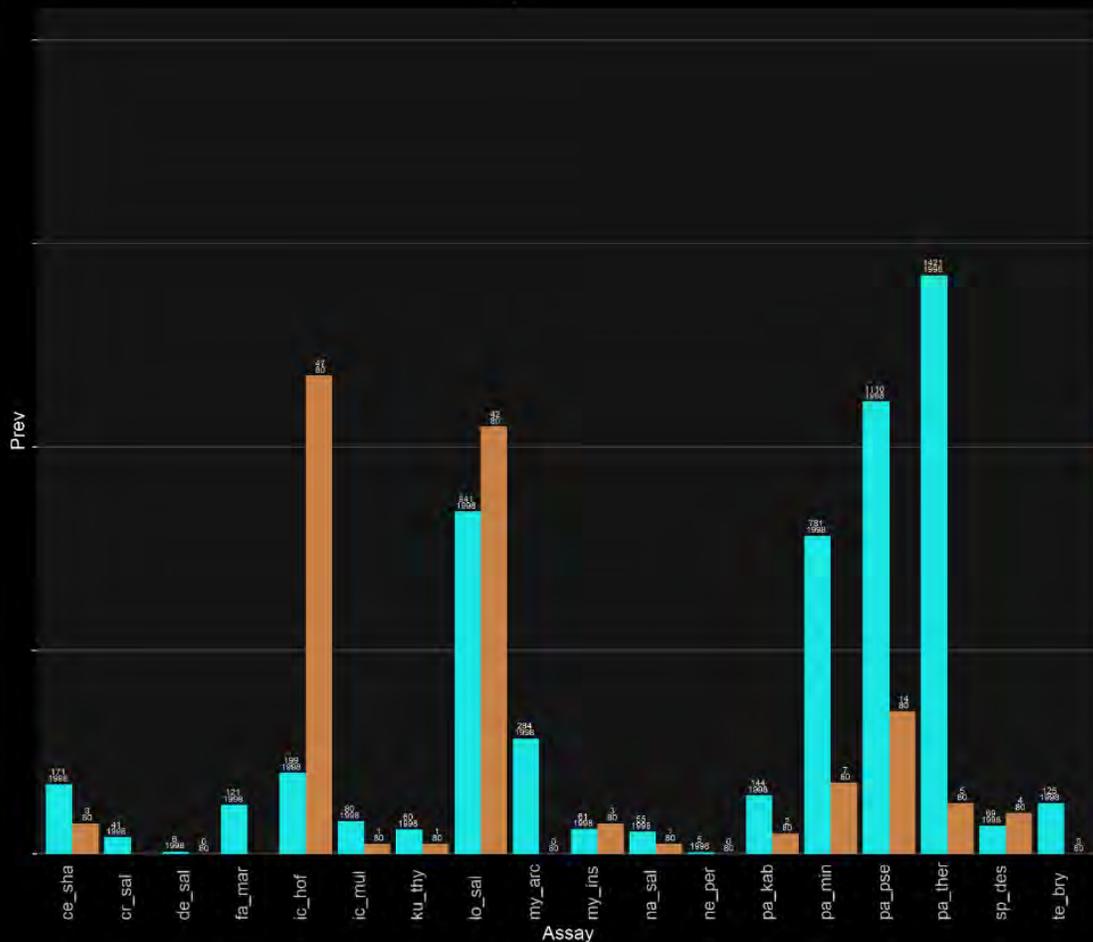


Coastal-GOA Bacterium Rel CT



*Candidatus Branchiomonas cysticola*  
Gill chlamydia

# GoA vs BC Coast: Coho - Parasites



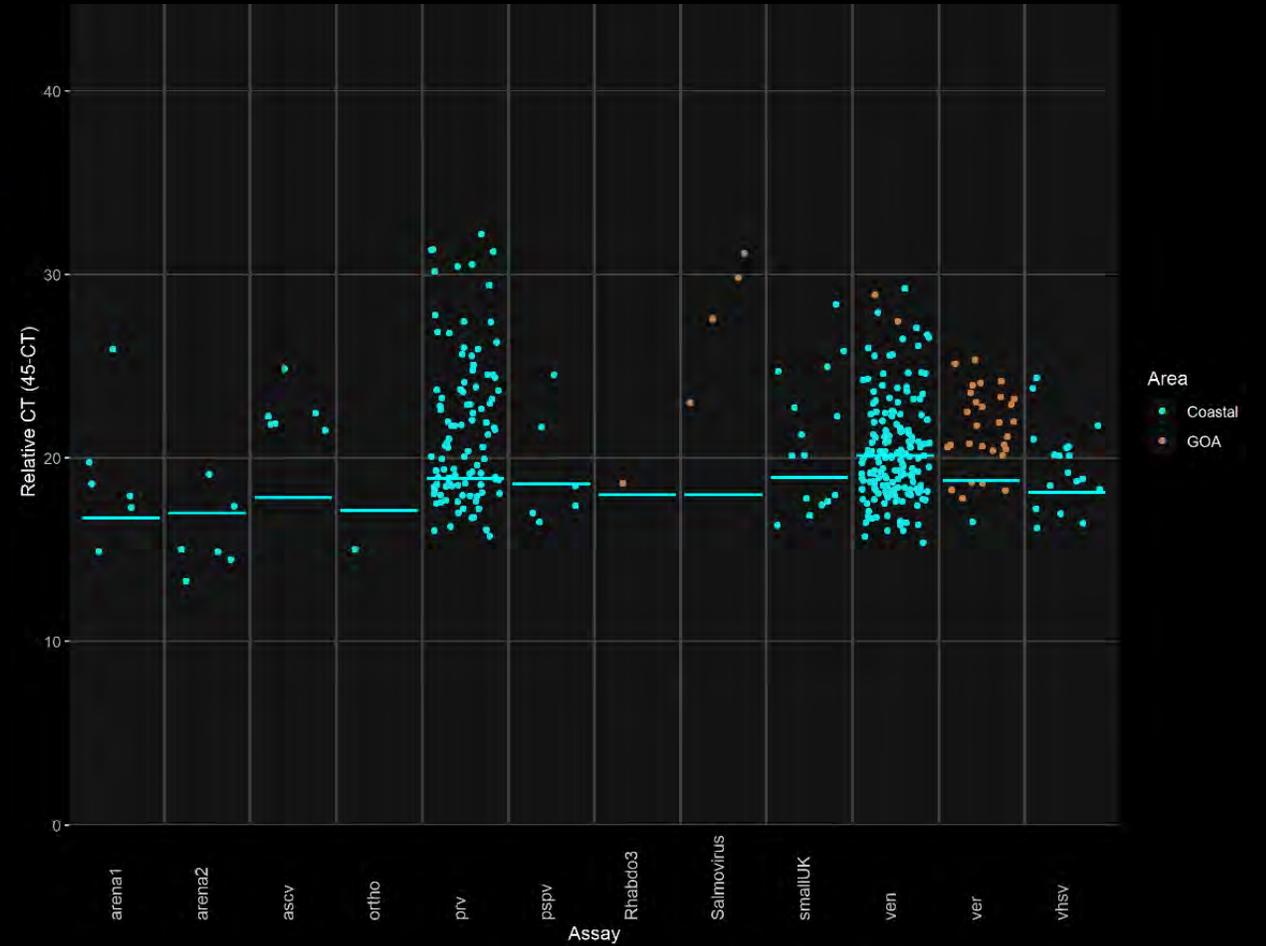
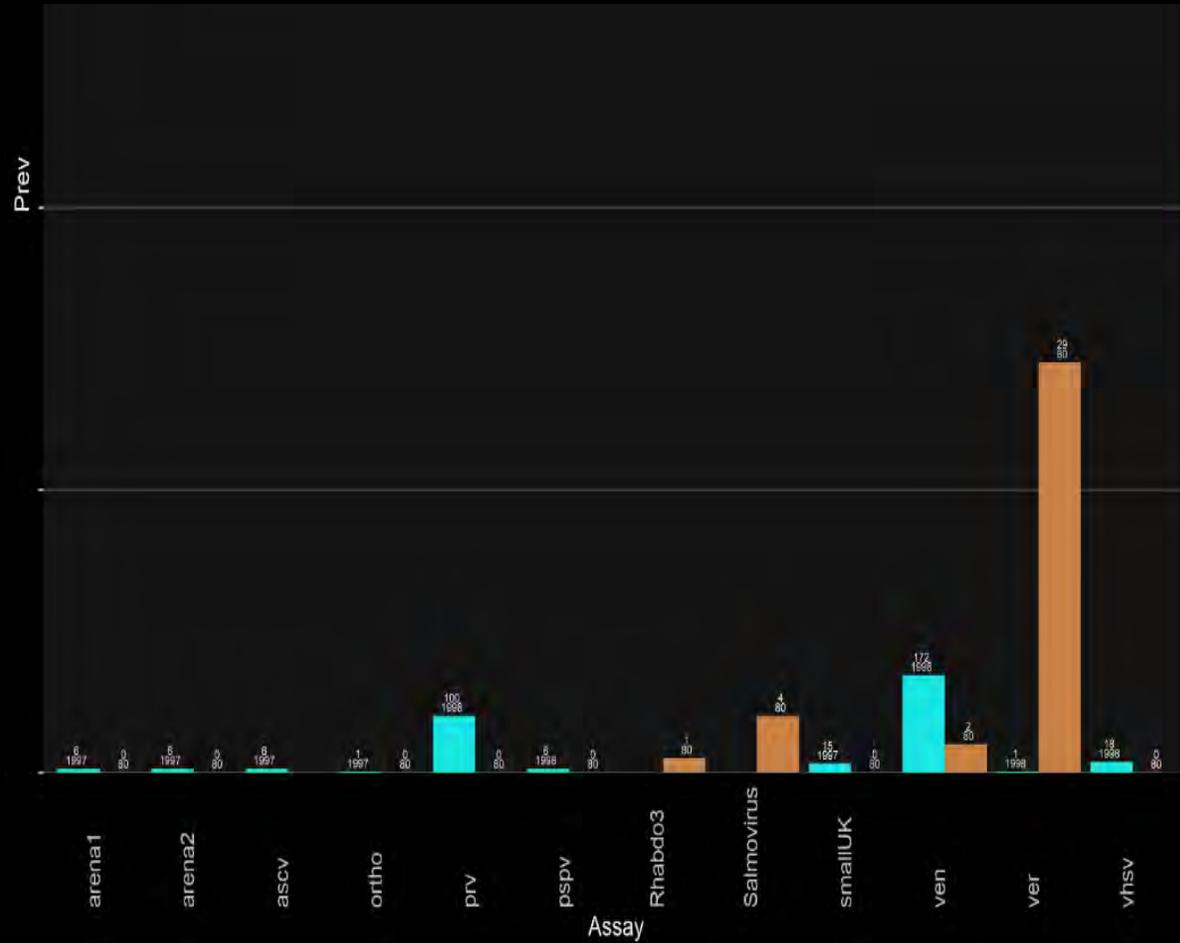
*Ichthyophonus hoferi*  
*Loma salmonae*  
*(Sphaerothecum destruens)*

FW:



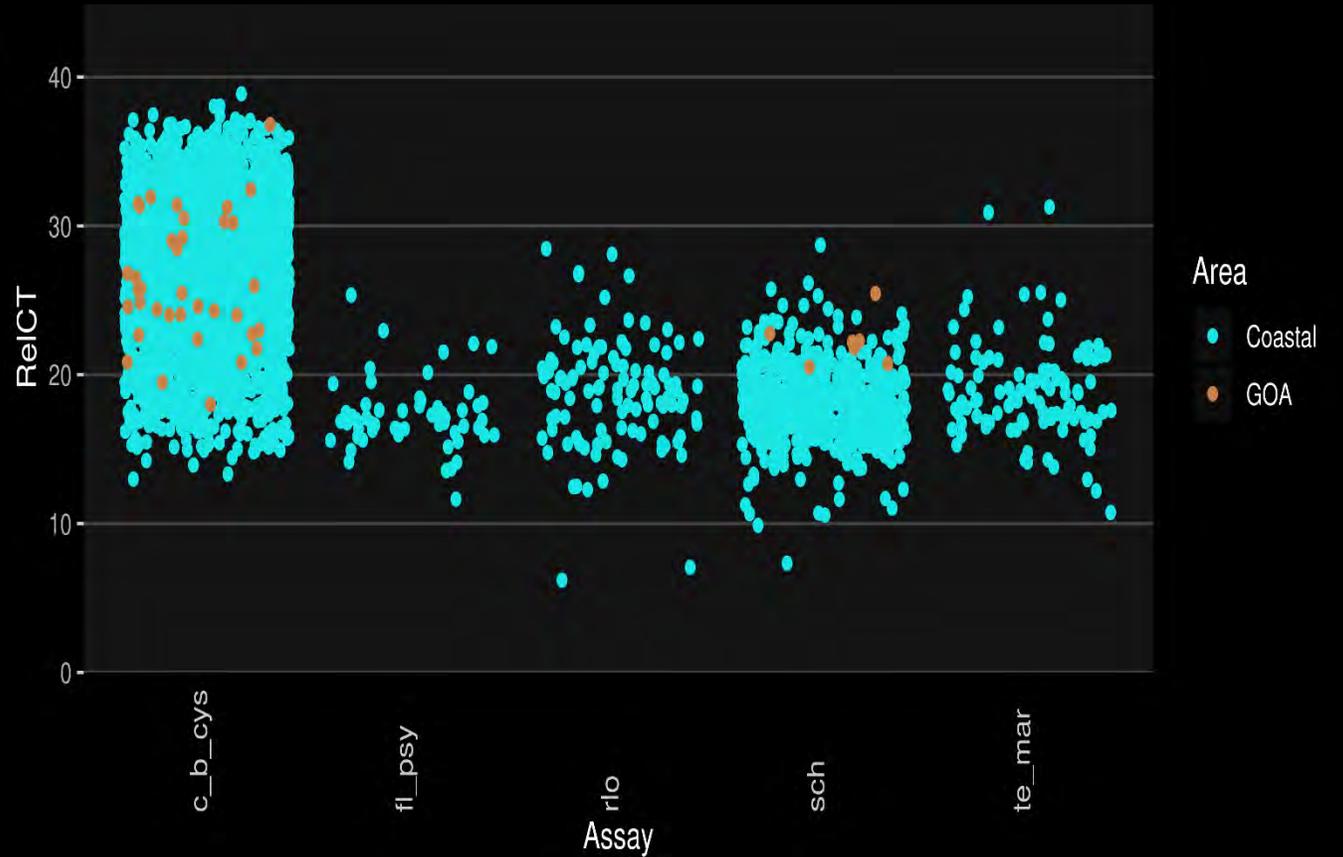
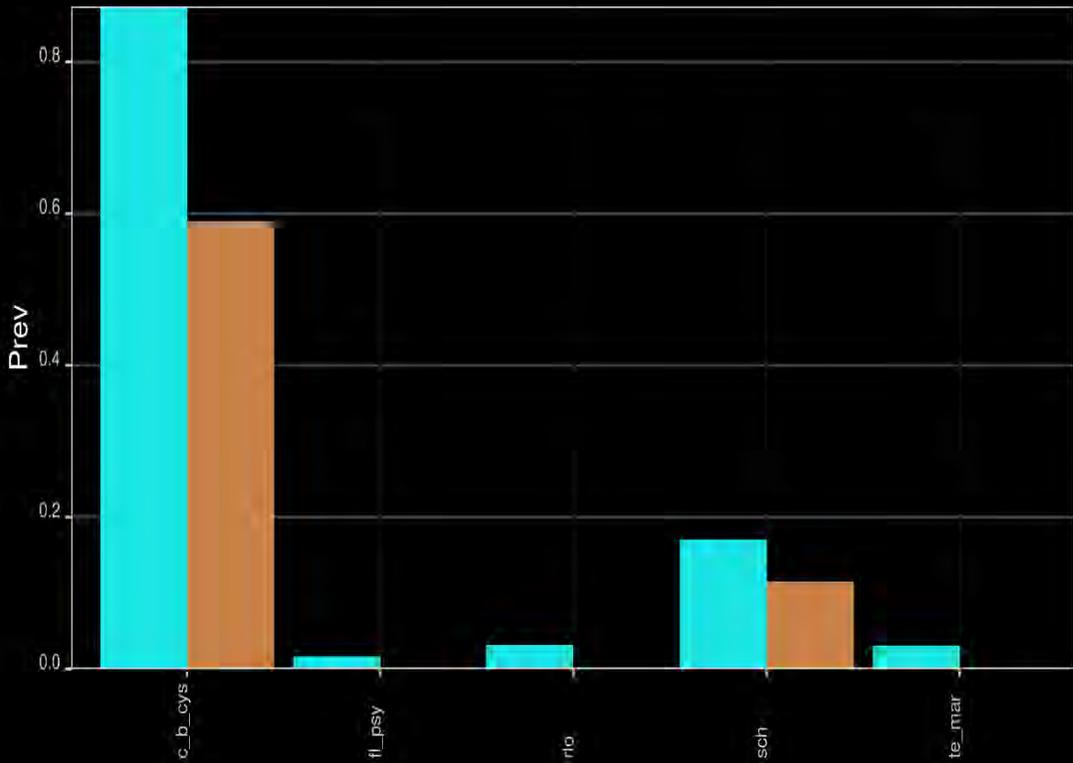
*Ceratonova Shasta*  
*Parvicapsula spp*  
*Myxobolus insidiosus*

# GoA vs BC Coast: Coho - Viruses



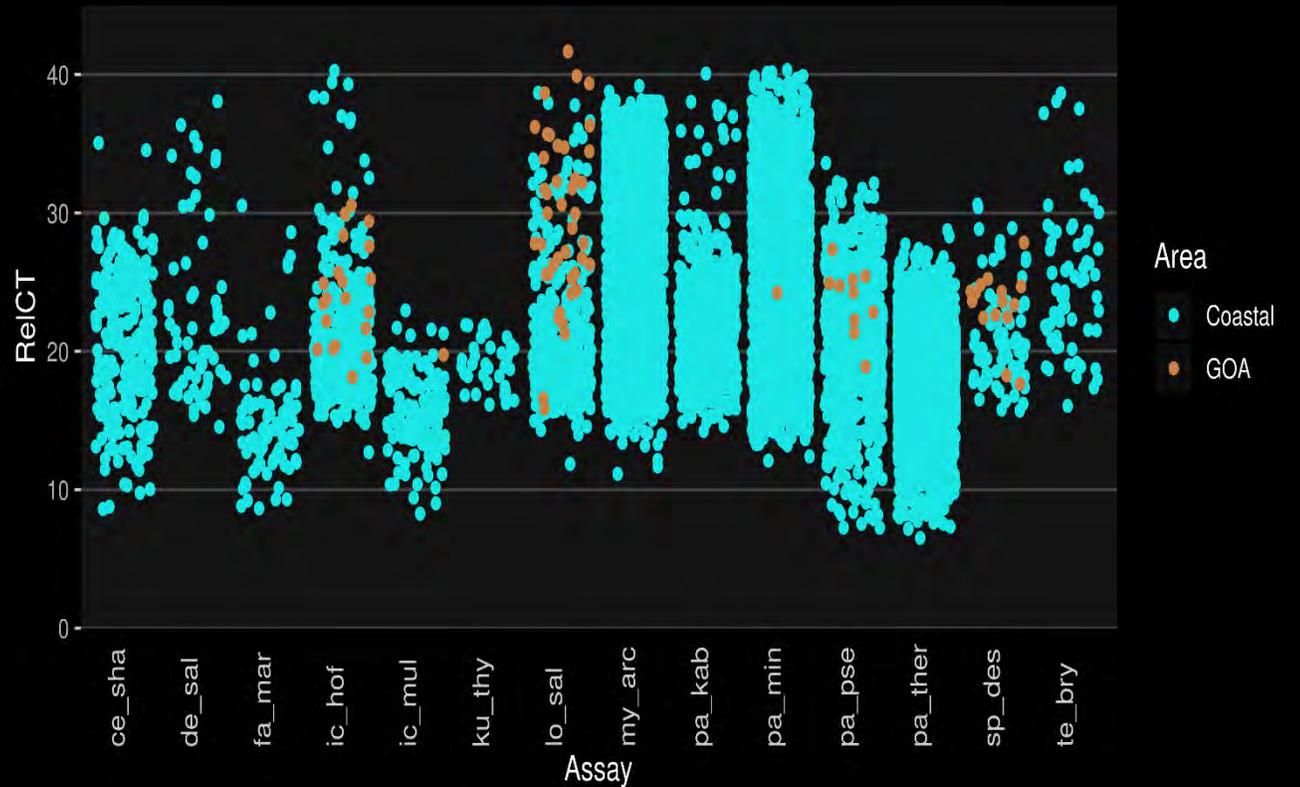
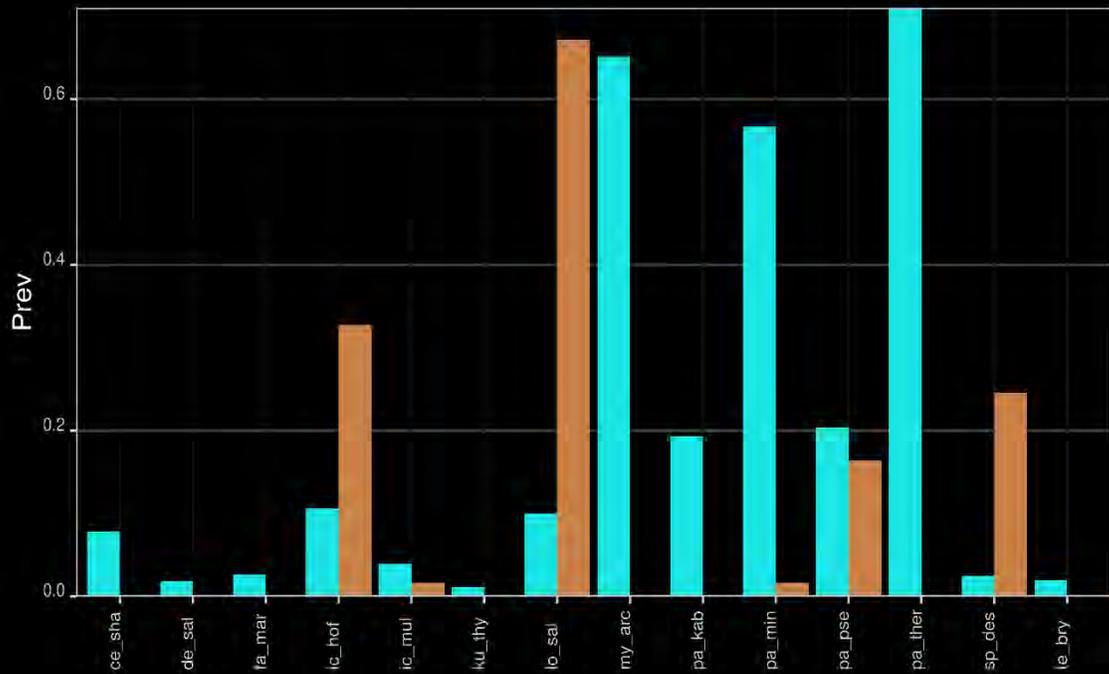

 Encephalopathy and retinopathy virus  
 (SalmovirusWFRC1\_virus)  
 (\*Rhabdo virus)

# GoA vs BC Coast: Sockeye - Bacteria



*C. B. cysticola*  
*Gill chlamydia*

# GoA vs BC Coast: Sockeye - Parasites

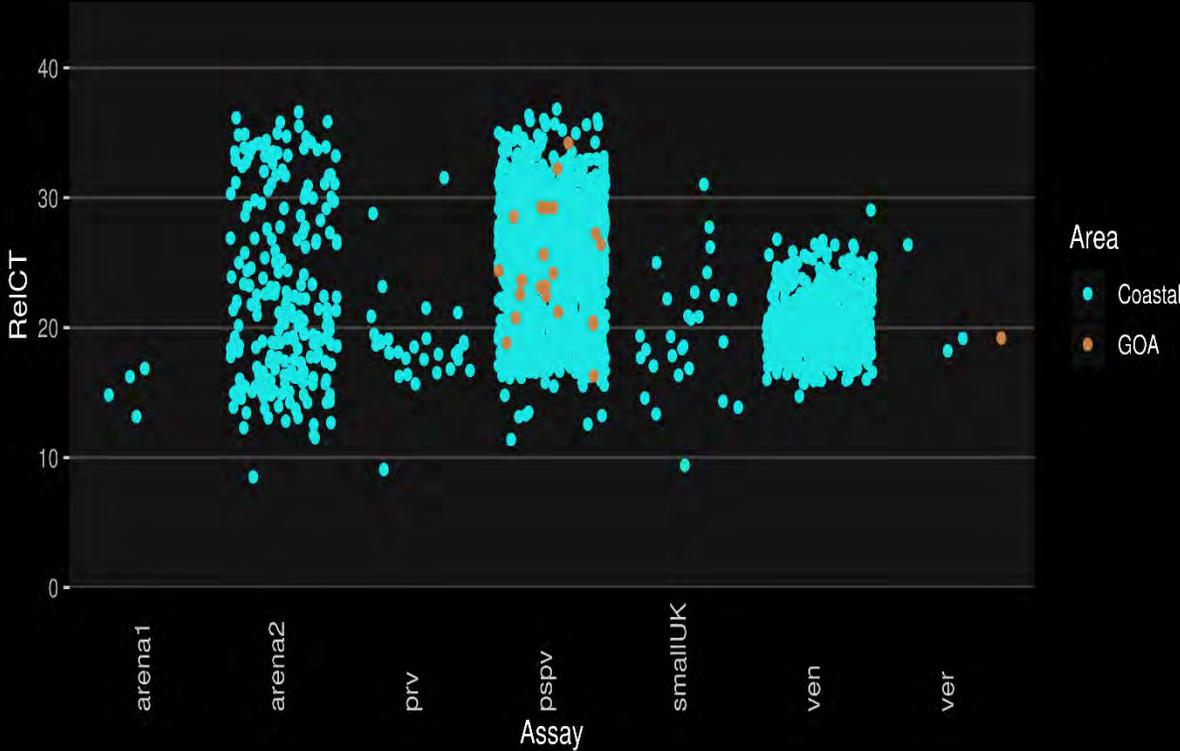
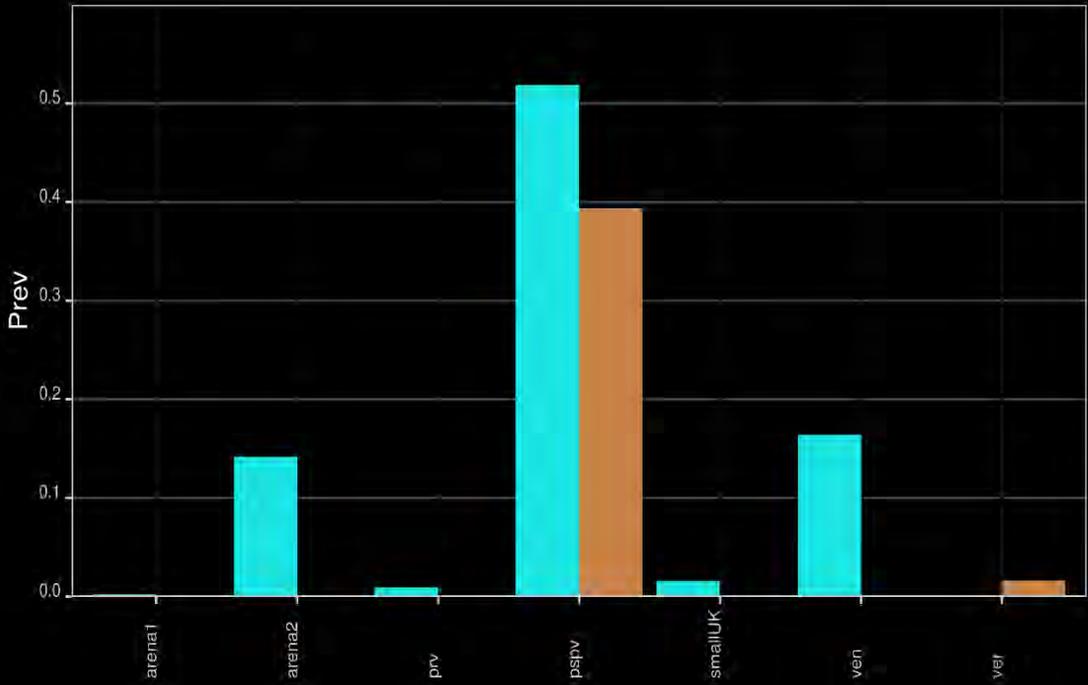



  
*Ichthyophonus hoferi*  
*Loma salmonae*  
*Sphaerothecum destruens*



*Parvicapsula pseudobranchicola*

# GoA vs BC Coast: Sockeye - Viruses

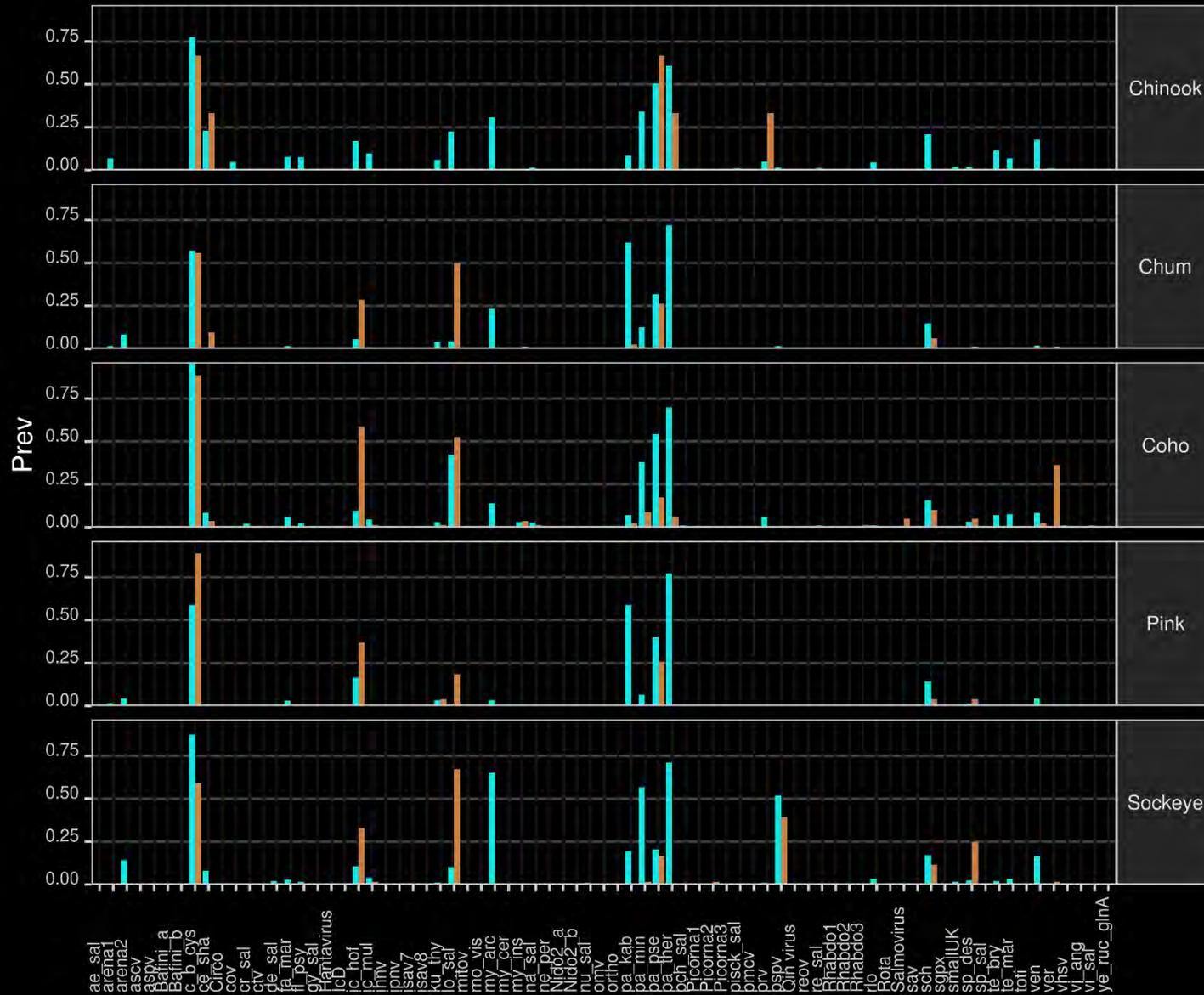


(Encephalopathy and retinopathy virus)



Pacific salmon parvovirus

# GoA vs BC Coast: All Species



- Coho and Sockeye trends recapitulated:

- *L. salmonae, I. hoferi,*

- Rising prevalence:

- Pink: *Candidatus Branchiomonas cysticola*

- Persistence

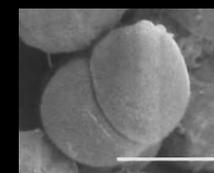
- *C. B. cysticola*

- *Parvicapsula pseudobranchicola*



# GoA vs BC Coast: Summary

- Most pathogens drop in prevalence
  - Not enough data for Chinook
- Rising prevalence:
  - *Ichthyophonus hoferi* : All species - Prey associated
  - *Loma salmonae*: All species - secondary infection?
  - Encephalopathy and retinopathy virus: Coho - Prey associated (piscivorous)
  - *Sphaerothecum destruens*: Sockeye
  - *C. B. cysticola*: Pink
- Persistence:
  - *C. B. cysticola*: All
  - *Parvicapsula pseudobranchicola*: All - Prey associated ?
  - Pacific salmon parvovirus: Sockeye
  - FW origin: *P. minibicornis* and *C. shasta*



# Summary

- GoA salmon have overall lower burden of pathogens than salmon sampled in their first fall/winter on the BC coast.
  - High mortality in early ocean phase / clearance of pathogens
  - Southern stocks from the US show a trend of higher pathogen prevalence and burden
- Pathogens of increasing prevalence are associated with lifestyle
  - Piscivorous diet!

# Future work

- More detailed analysis
  - RIB comparison for all species
  - Correlation with condition and observations
  - Stock specific analysis once they become available
- Fit chip data
  - Correlate disease and stress with condition and stock
    - *I. hoferi* -> osmoregulation
  - Include Ichthyobodo
- Histopathology to determine disease potential of fish with high pathogen loads
  - *I. hoferi*
- **eDNA**



**Prof. Kaganovsky crew**

**IYS team:**

Alexey Somov,

Albina Kanzeparova,

Svetlana Esenkulova,

# Thank you!

**Molecular  
Genetics Lab:**

Karia Kaukinen,

Tobi Ming, Amy

Tabata, Angela

Schulze, Shaorong

Li, Emiliano Di

Cicco

Richard Beamish



INTERNATIONAL  
YEAR OF THE SALMON