## Conditions that Promote or Inhibit Off-flavor in RAS



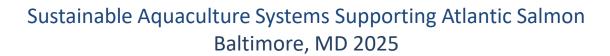


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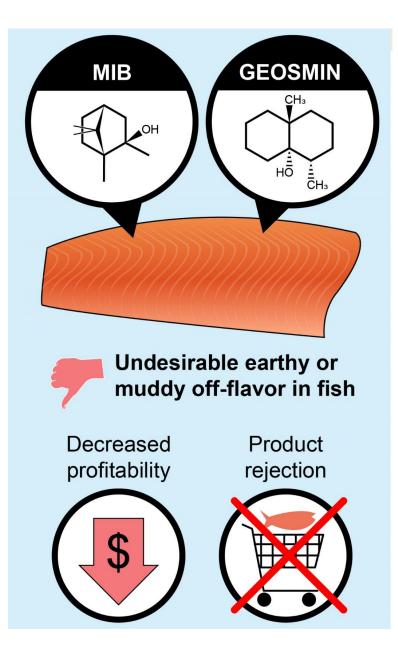






Bacteria produce Geosmin & MIB in RAS.



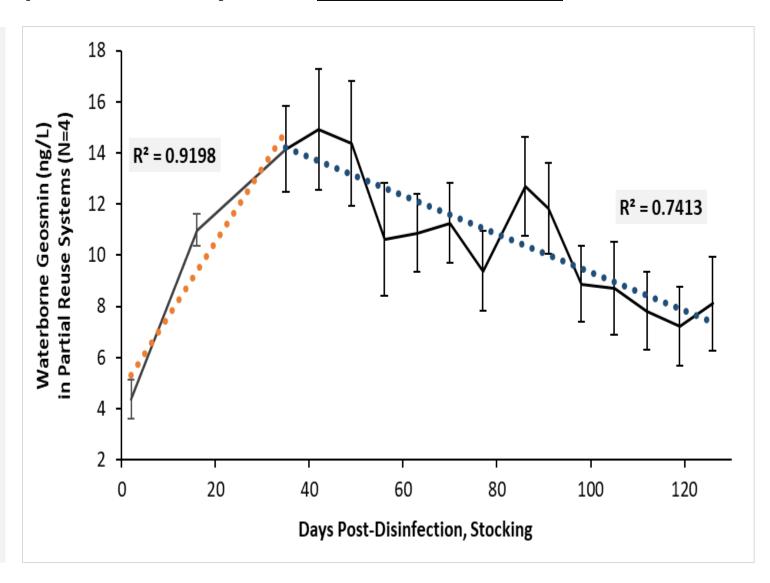


- > Depuration is a proven finishing procedure.
  - Fish are moved to separate systems.
  - Water low/void of off-flavor is exchanged.
  - Off-flavor is eliminated by fish across a concentration gradient with the water.

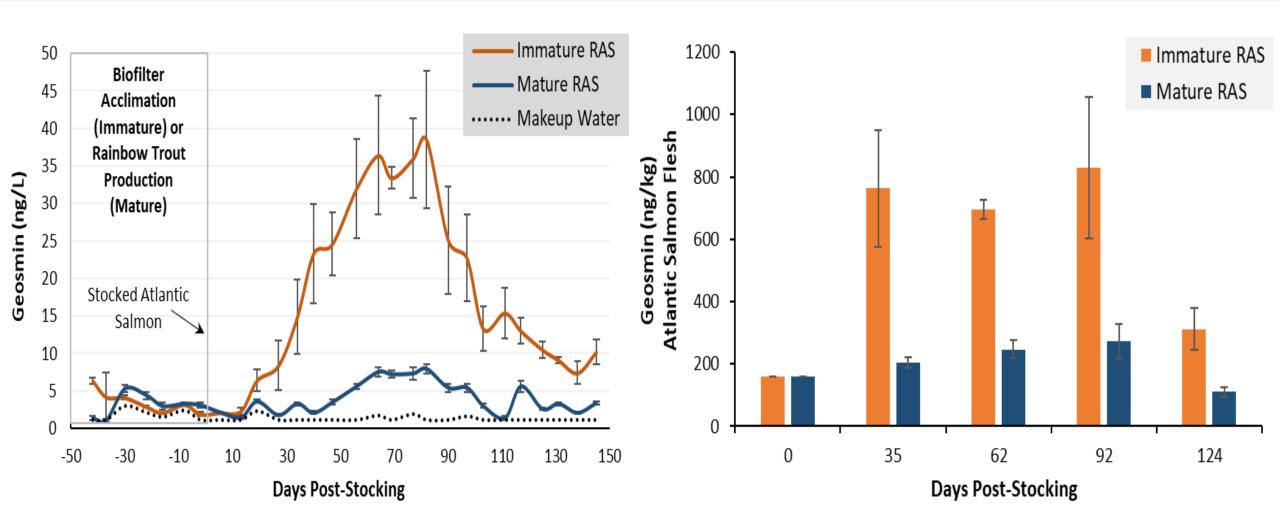


#### Geosmin production in partial reuse systems without a biofilter.

- This data set was the driver for subsequent studies at FI.
- ➤ We cleaned and disinfected 4 identical partial reuse systems (PRS), then stocked Atlantic salmon. We did not brush submerged surfaces thereafter.
- ➤ A peak and gradual decline in waterborne geosmin was observed across replicate PRS.
- ➤ Does a similar response occur in RAS?
  - Is it related to the development of microbial maturity?

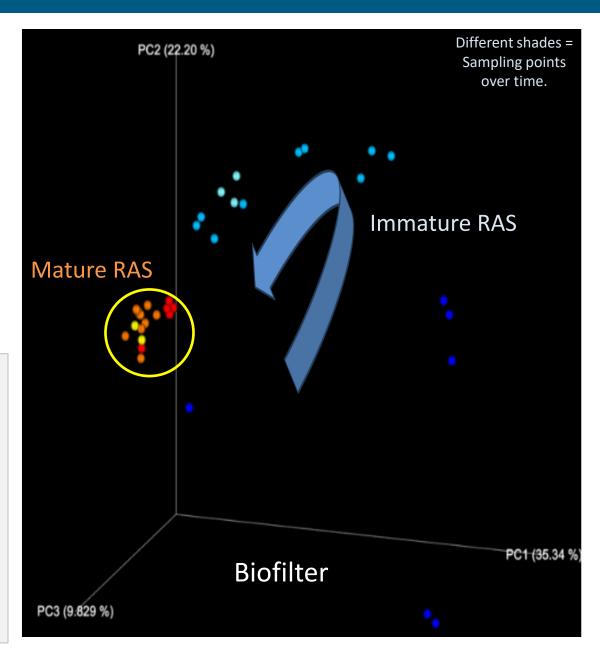


- ➤ A similar response was observed in replicated RAS (n=3).
- At its peak, geosmin was 4x higher in water and Atlantic salmon flesh in pre-disinfected RAS where nitrification was re-established (*Immature*) vs. RAS operated for 2.5 years (*Mature*).



Off-flavor Producing Bacteria	Off-flavor Degrading Bacteria
Odorella benthonica	Pseudomonas putida
Calothrix sp. PCC7507	Stenotrophomonas maltophilia
Cylindrospermum stagnale	Pseudomonas fluorescens
Synechoccus sp. NIES-970	
Sorangium cellulosum	
Aquabacterium commune	

- Four of six off-flavor (OF) producers were Cyanobacteria.
- ➤ Most OF producers were found in tank biofilm.
- ➤ OF producer abundance was generally <u>higher in the</u> mature RAS, opposite to the OF levels.
- ➤ The plot at right is a principal coordinates depiction of the entire microbiome over time in biofilters.



- > Despite similar water flushing and feeding, many water quality differences were observed.
  - Two machine learning models separated true color as the most important variable affecting geosmin.
- > The decline in OF production coincided with stabilized feeding and nutrient levels, and improved nitrification.

# Conditions including Significant WQ Differences Associated with Higher Off-flavor

**Recently Disinfected** 

Immature, Less Stable Microbiome

Higher Total Ammonia Nitrogen

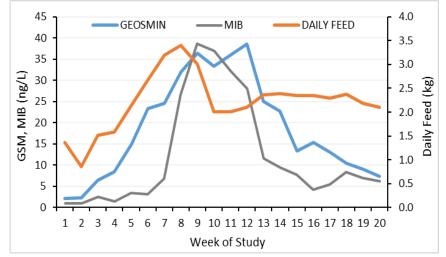
Higher Nitrite Nitrogen

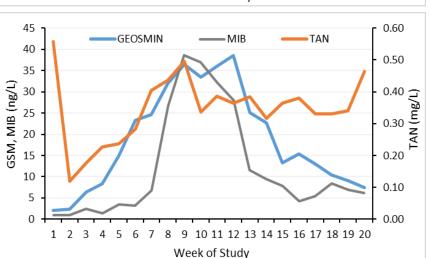
**Higher True Color** 

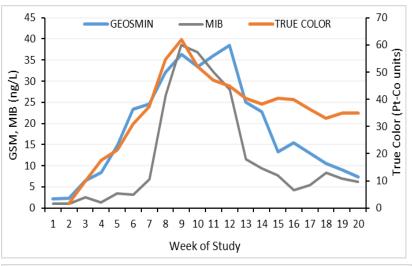
**Higher Total Suspended Solids** 

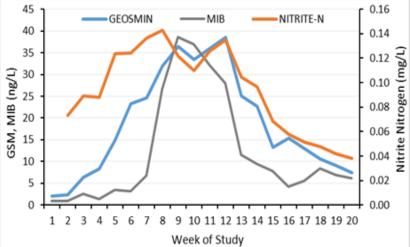
**Higher Heterotrophic Bacteria Counts** 

Lower Nitrifier Abundance









## CONSERVATION FUND

### Can Common Oxidants Control Off-flavor?

> Hypothesis 1: Low-dose ozone will create an environment conducive to inhibiting off-flavor production in RAS?

Conditions Associated with Lower Off-flavor	Ozone Control
Longterm RAS Operation	-
Mature & Stable Microbiome	-
Lower Total Ammonia Nitrogen	٧
Lower Nitrite Nitrogen	$\downarrow$
Lower True Color	$\downarrow$
Lower Total Suspended Solids	$\downarrow$
Lower Heterotrophic Bacteria	$\downarrow$
Higher Nitrifier Abundance	<b>フ</b> ピ

Hypothesis 2: Low-dose ozone with peracetic acid dosing will reduce off-flavor levels through advanced oxidation or combined water quality control and microbial reduction.



- > Operating RAS continuously without shutdown maintains lower offflavor levels vs. disinfecting and restarting between cohorts.
- ➤ Better water quality, including lower true color (dissolved organics), and lower TAN and nitrite resulting from stable and efficient nitrification, aligned with lower off-flavor levels in water and fish.
- > We must consider off-flavor production holistically.
  - Geosmin and MIB are produced by specific bacteria *competing for space* and nutrients in a complex microbiome.
- This research is a step forward in establishing a possible "recipe" for maintaining lower geosmin and MIB levels in RAS.
  - Optimizing RAS operation, water treatment efficiencies, and the RAS environment could be a cost-effective solution!





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