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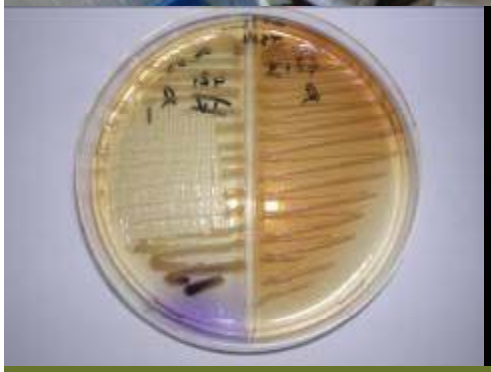
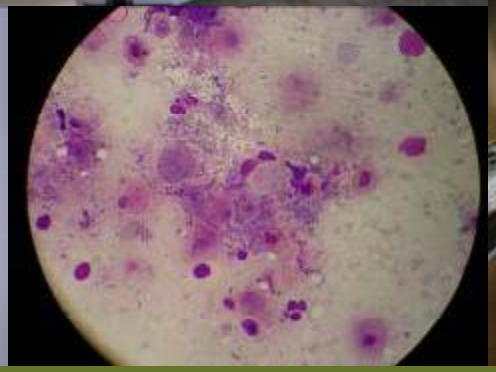
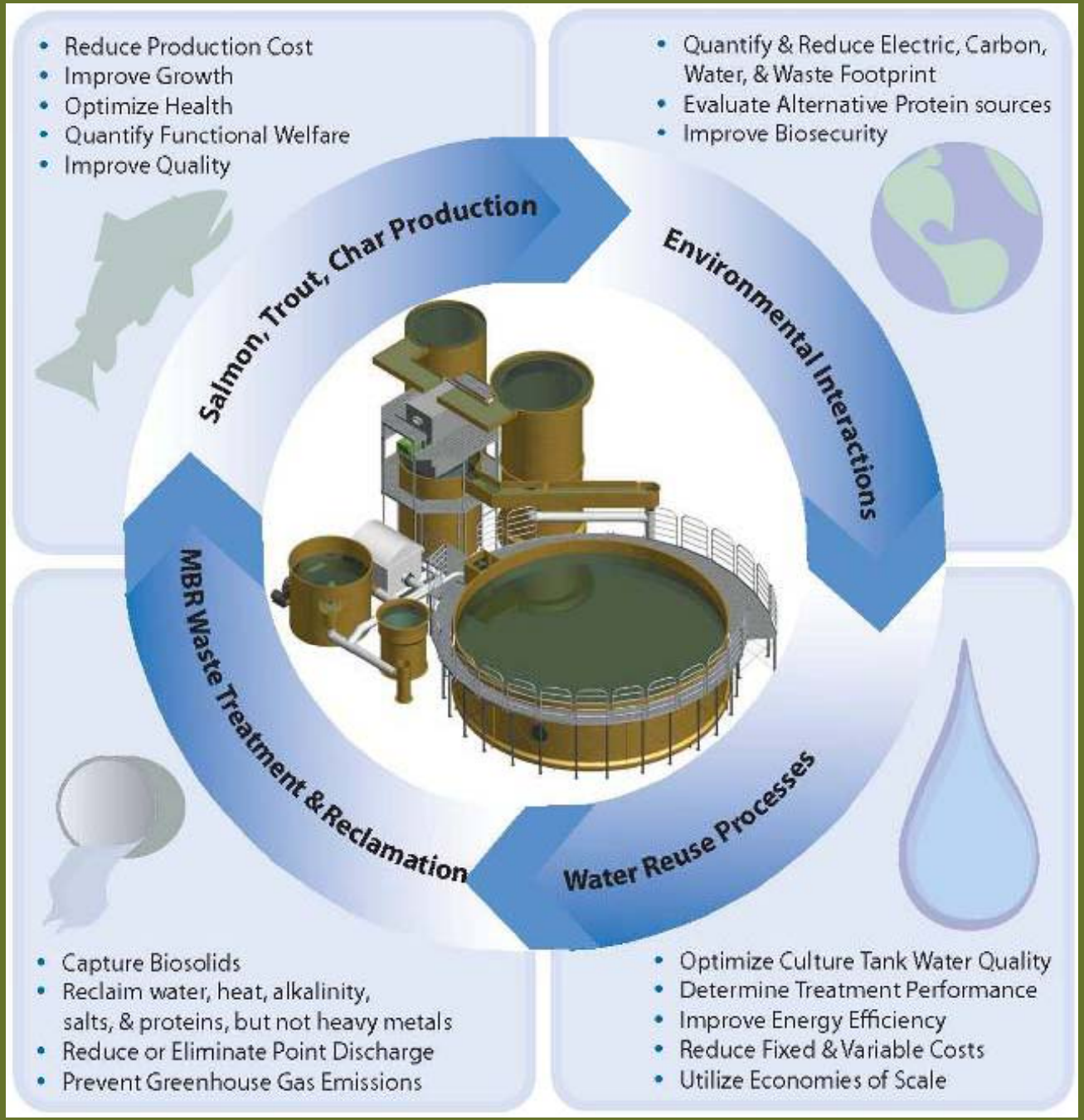
Biosecurity in Water Recirculation Aquaculture Systems





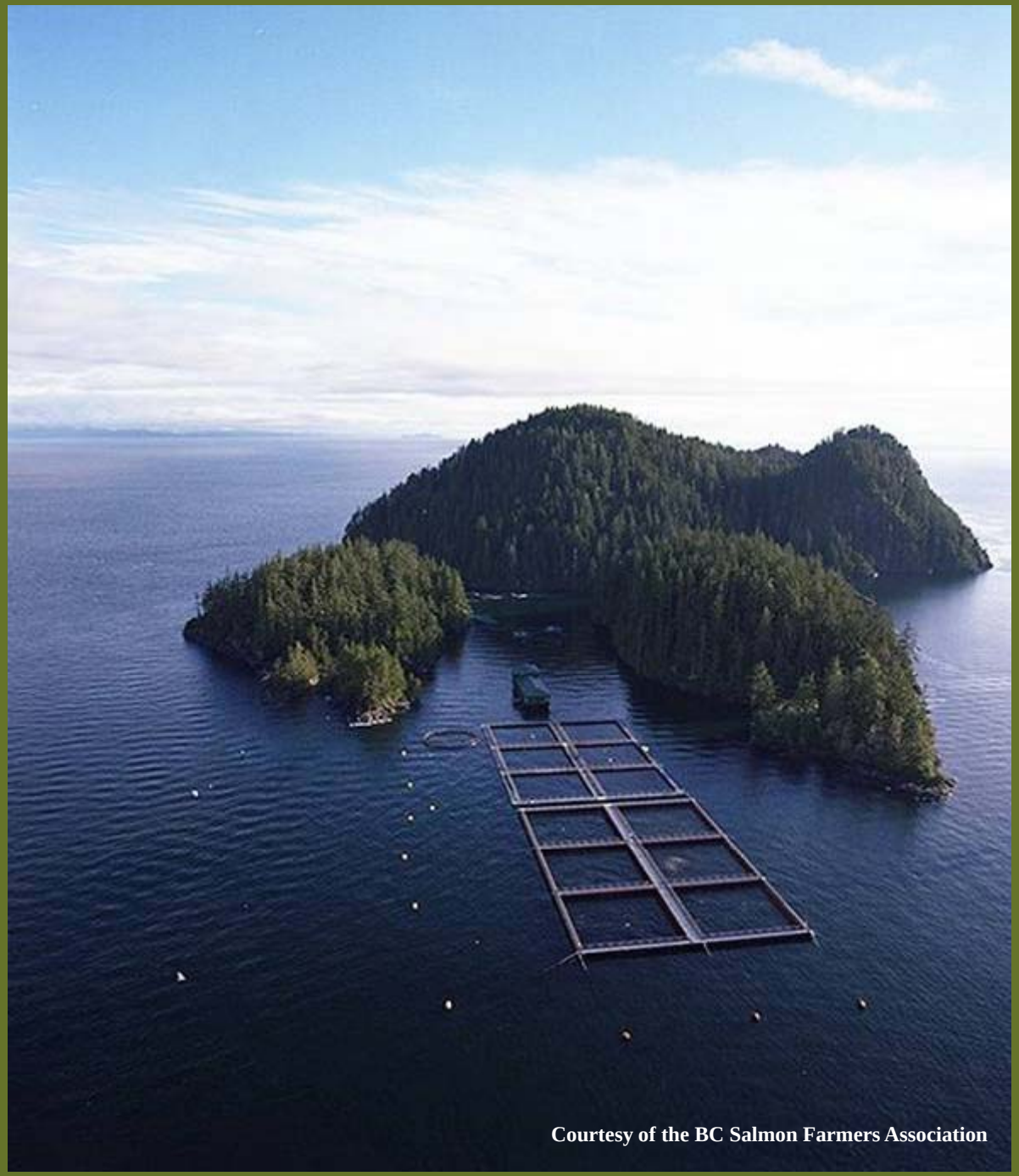
Christopher Good

Biosafety and Biocontainment Symposium
Baltimore, Maryland
February 6-9



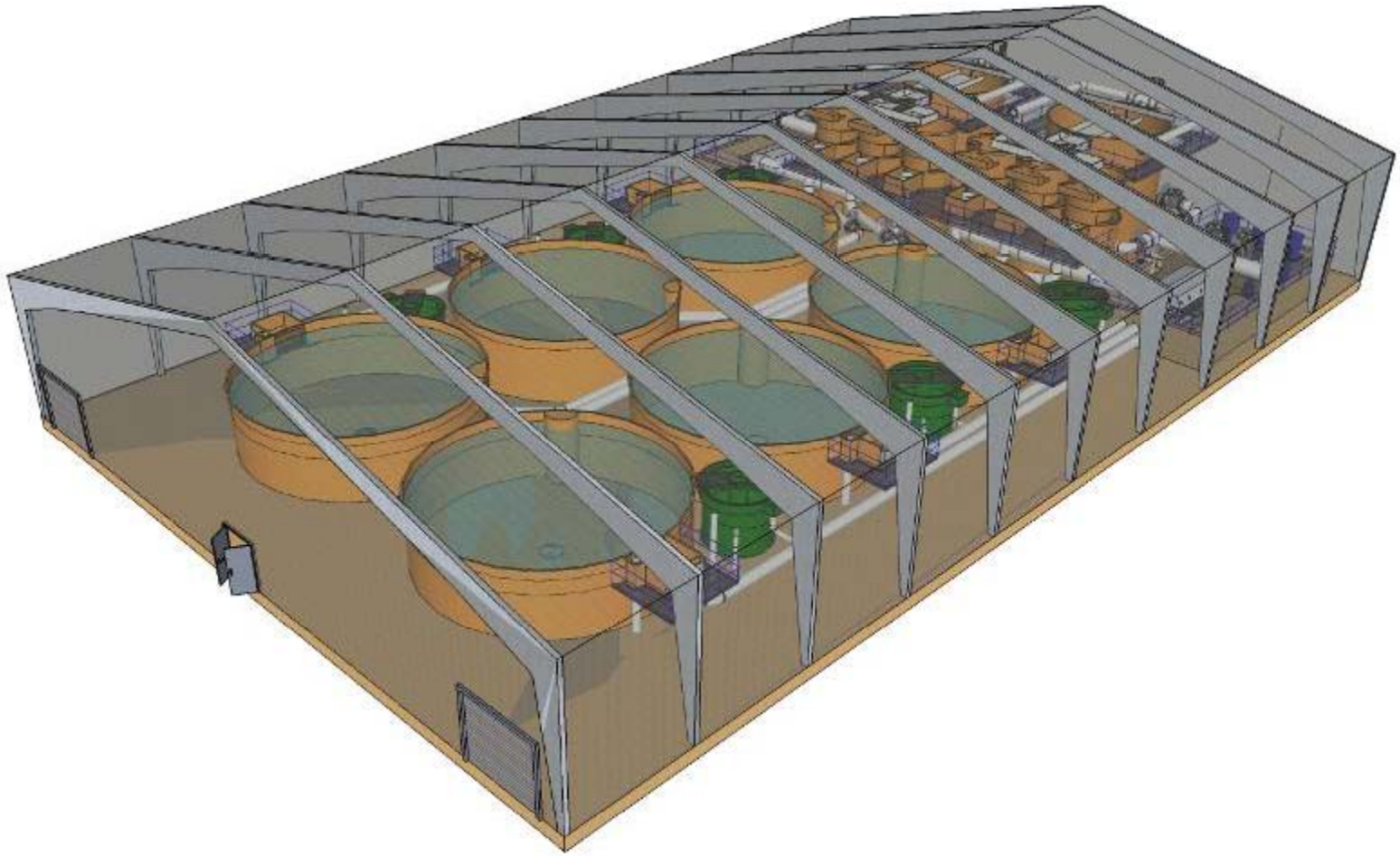
Research at The Freshwater Institute

At Issue...

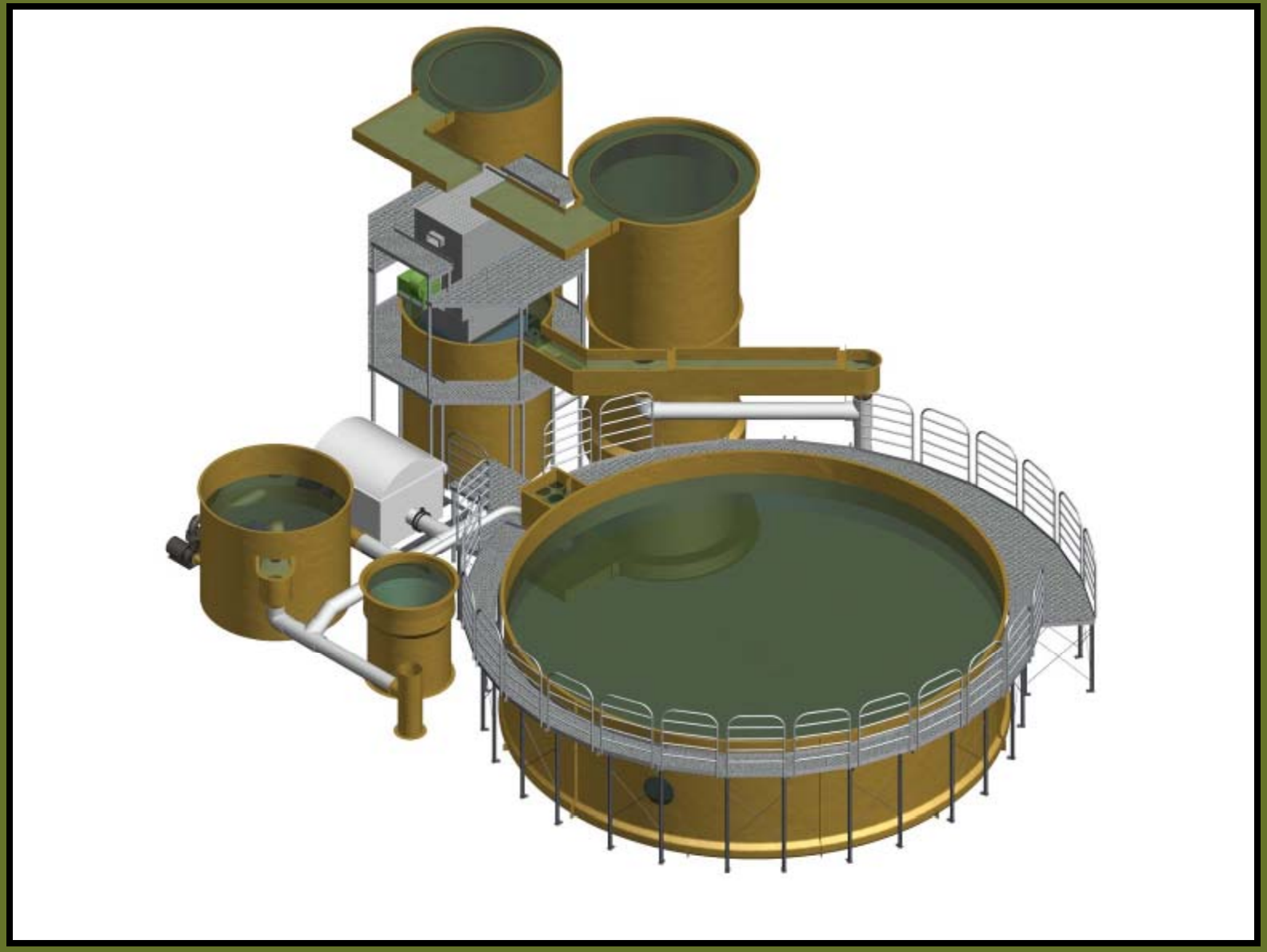


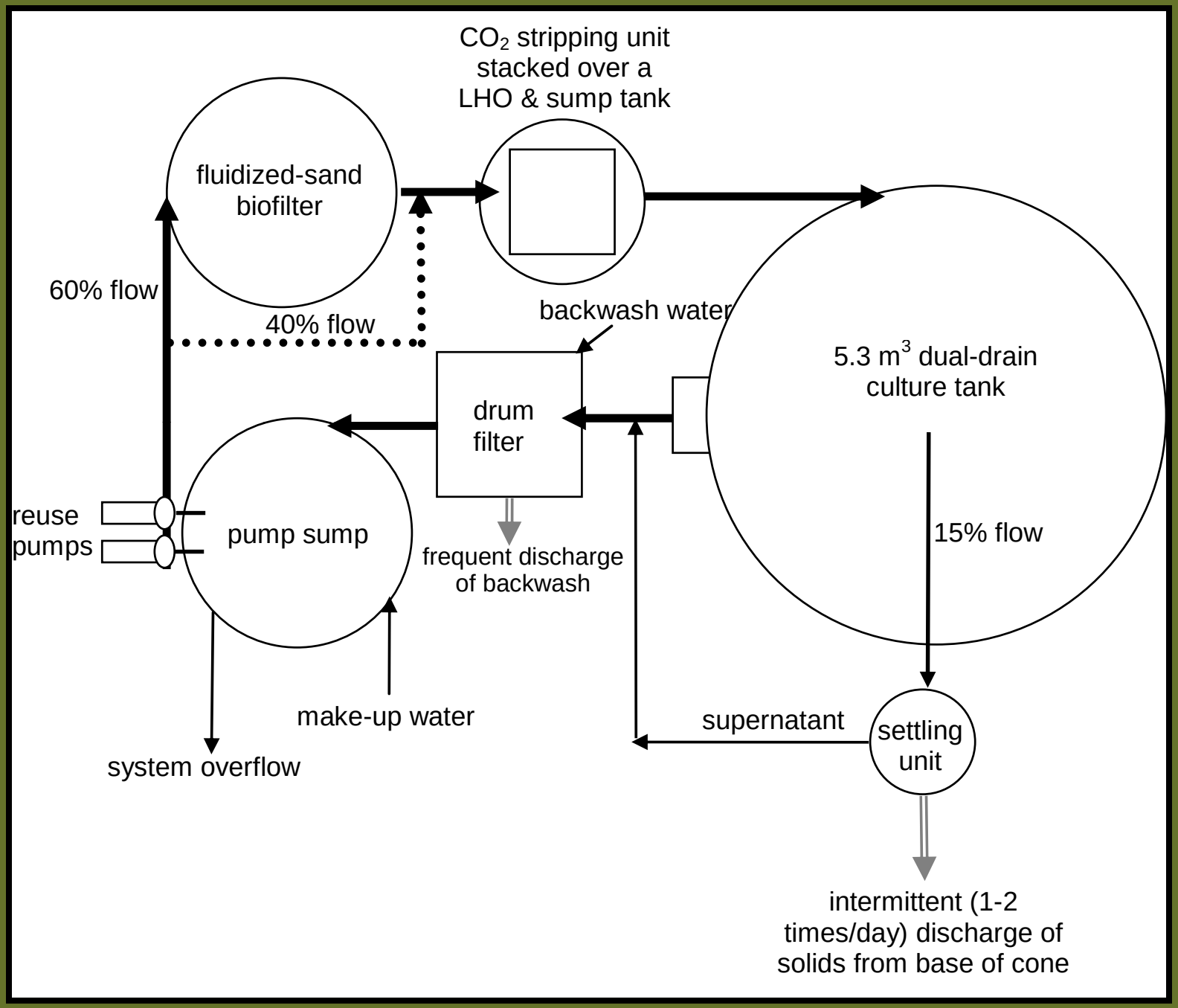
Courtesy of the BC Salmon Farmers Association

Closed Containment Facilities with Water Recirculation



Freshwater Institute
Commercial Scale Recirc System




CO_2 stripping unit
stacked over a
LHO & sump tank
fluidized-sand
biofilter
60% flow
40% flow
backwash water
5.3 m^3 dual-drain
culture tank
drum
filter
reuse
pumps
pump sump
frequent discharge
of backwash
15% flow
make-up water
supernatant
settling
unit
system overflow
intermittent (1-2
times/day) discharge of
solids from base of cone

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Prevent Pathogen Introduction via **Water**

- Use **ground water**
  - Avoids obligate pathogens, but presence of opportunists must always be assumed
- Capture ground water before it becomes contaminated
- Spring head ponds can harbor fish and attract birds

Prevent Pathogen Introduction via **Water**

Prevent Pathogen Introduction via **Eggs**

- **Use certified pathogens free eggs when possible**
  - **surface disinfect eggs upon arrival**

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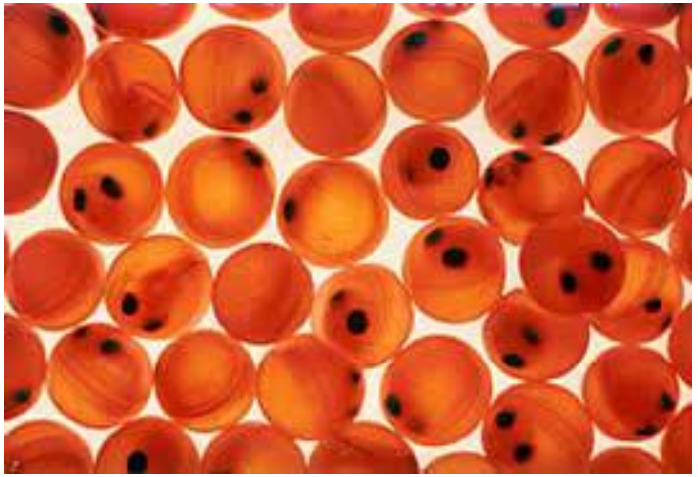
Advantages of Closed Containment

With biosecure source water and specific pathogen-free eggs:

- **No vaccinations required**
- **Reduction or elimination of antibiotic and chemotherapeutant usage**



Advantages of Closed Containment

Avoids <u>obligate pathogens</u>:

- **Viruses**
  - **IPNV**
  - **IHNV**
  - **VHSV**
- **Bacteria**
  - ***Aeromonas salmonicida***
  - ***Renibacterium salmoninarum***
- **Parasites**
  - ***Ichthyophthirius multifiliis***
  - ***Myxobolus cerebralis***

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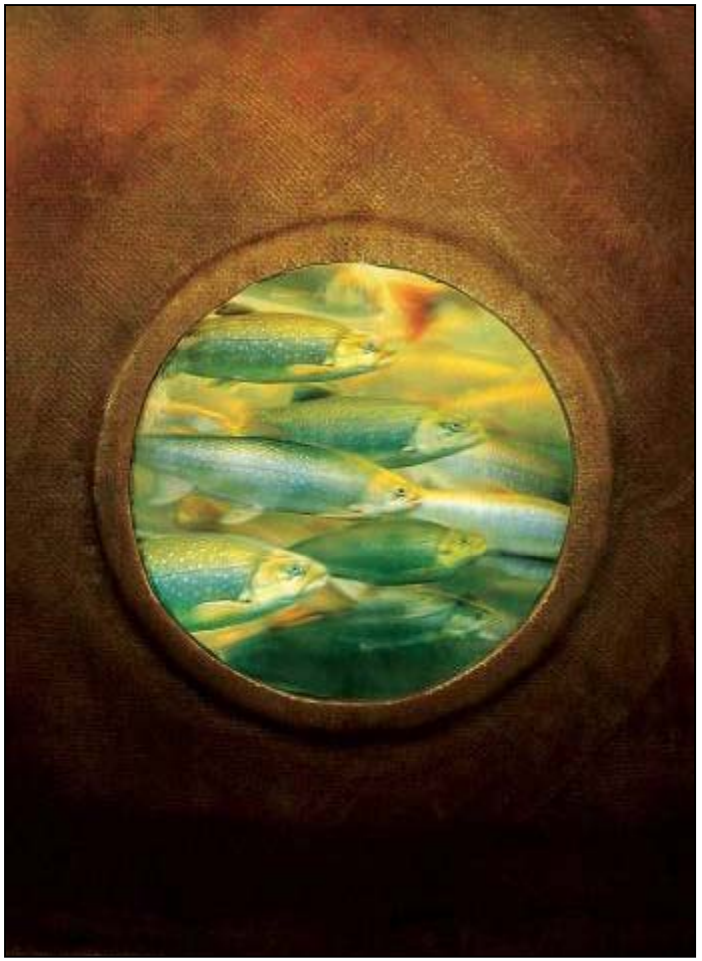
Advantages of Closed Containment

<u>Opportunistic pathogens</u> still need to be considered

Closed containment allows for excellent **<u>control of environmental conditions</u>** to prevent opportunists from gaining "upper hand"



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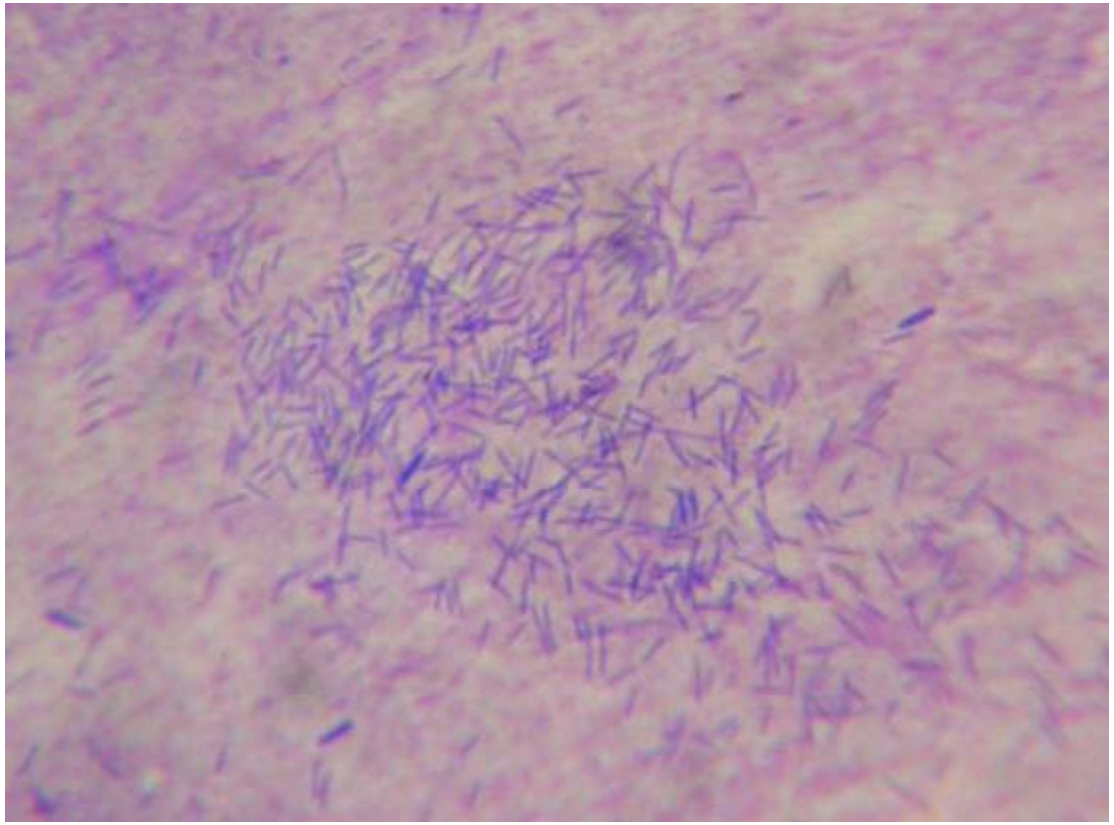
Flavobacteria complex of fish pathogens

- Long, filamentous Gram –ve rods
- Ubiquitous in the freshwater environment
- Cause disease during unfavorable environmental conditions



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Flavobacteria complex of fish pathogens

Flavobacterium psychrophilum

coldwater disease

Flavobacterium columnare

columnaris

Flavobacterium branchiophilum

bacterial gill disease (BGD)

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F. psychrophilum
Coldwater disease

- Outbreaks tend to occur at 4-10ºC
- Most severe in 0.5-5 gram fish
- Lesions often begin at the adipose fin and progress to the entire caudal peduncle

http://www.lsc.usgs.gov/fhb/leaflets/FHB86.pdf

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F. psychrophilum
Coldwater disease

- Integument and musculature necrosis
- Virulence factors include adhesins, exotoxins, and proteases
- Spinal deformities can occur in survivors
- Horizontal and vertical transmission

http://www.lsc.usgs.gov/fhb/leaflets/FHB86.pdf

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F. columnare columnaris

- Cloudy, fungus-like patches, often around dorsal fin
- Progress to ulcerative, necrotic lesions
- Lesions sometimes bright yellow

www.umesc.usgs.gov

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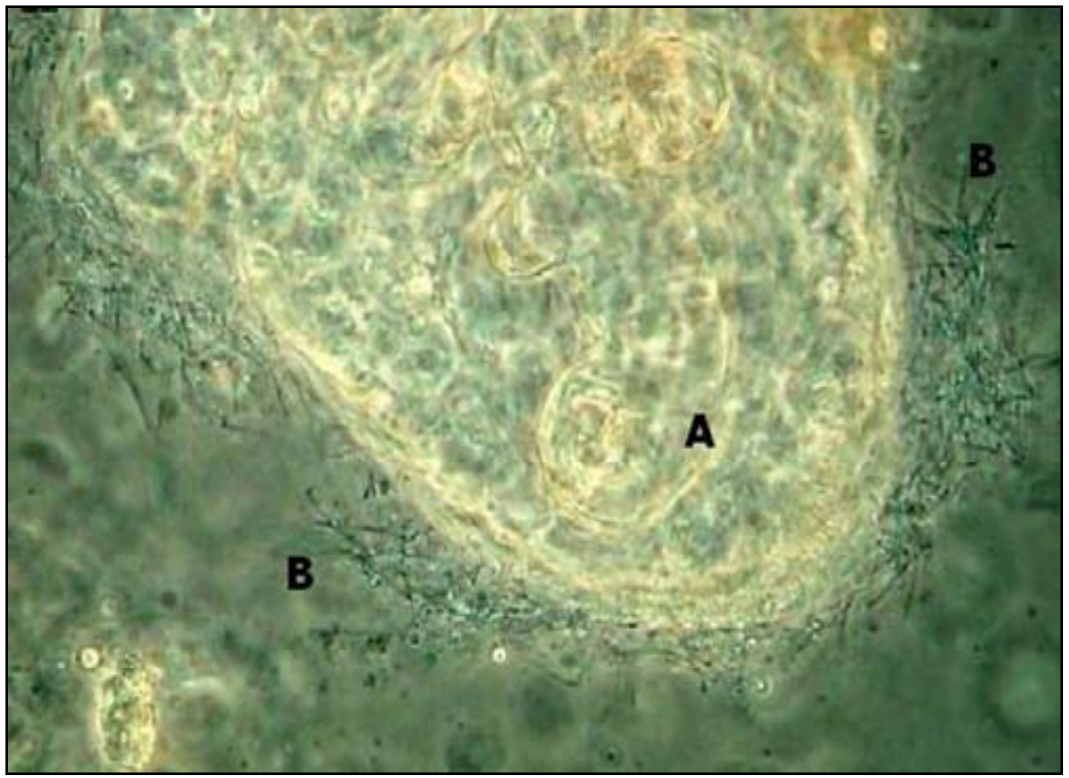
F. branchiophilum
Bacterial Gill Disease

- External infection of the gills – respiratory distress
- Outbreaks in younger fish can be devastating
- Need to treat **early** – Perox-Aid FDA approved for salmonids
- Take fish off feed



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F. branchiophilum
Bacterial Gill Disease

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F. branchiophilum
Bacterial Gill Disease

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Protozoan Parasites

Ichthyobodo (costia)

Chilodonella

Epistylis

Trichodina

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Saprolegnia spp.

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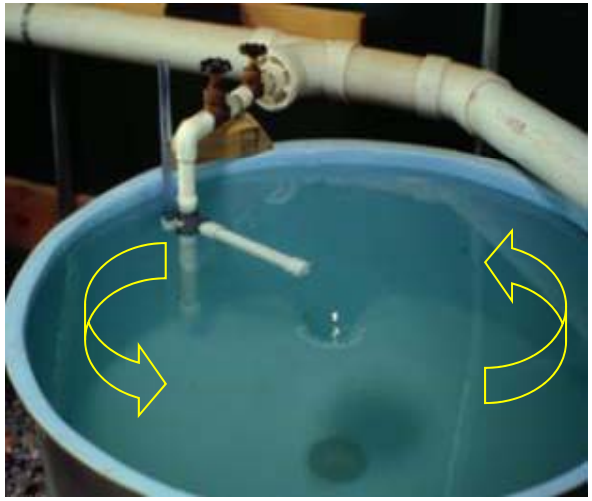
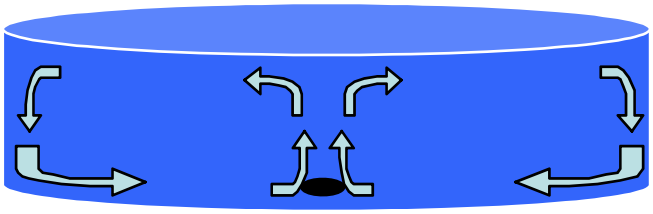
Environmental Management: Circular Tanks

- Tank hydraulics can be optimized
- Solids flushing can be very rapid, < 1-3 min
  - Tanks are self-cleaning
  - Fresh fecal pellets are flushed before they breakdown
  - Solids are not stored in the culture tank primary flow



primary rotating flow

secondary radial flow



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Environmental Management: Circular Tanks

Outcomes:

Excellent
solids control
and
uniform water quality
throughout tanks

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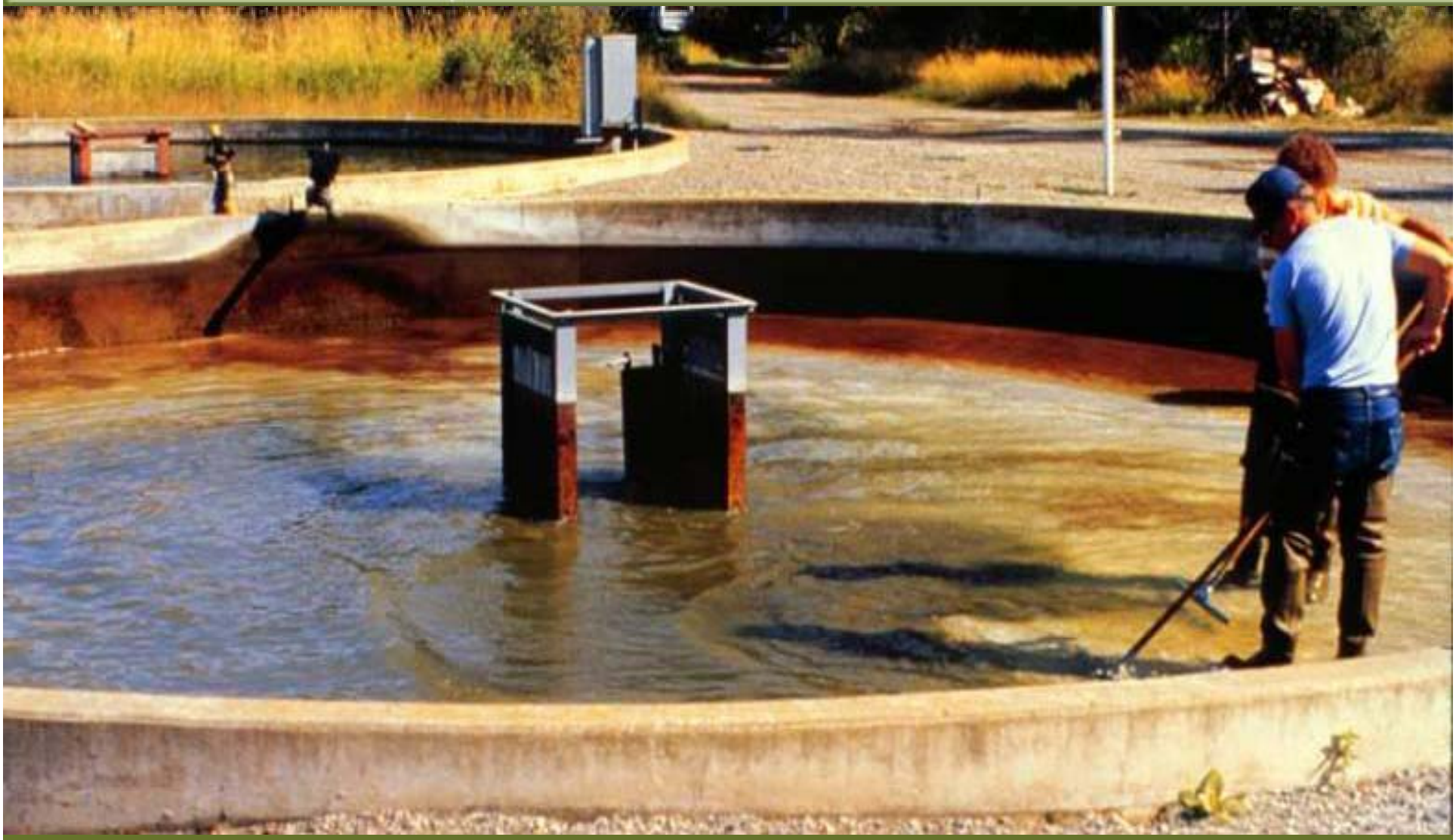
* Circular Tanks Without Rotation *



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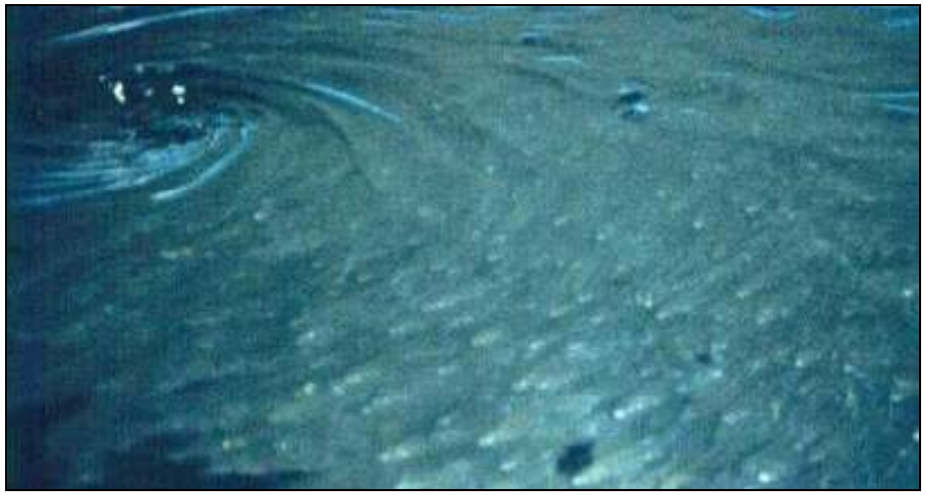
Environmental Management: Optimum Swimming Velocity

- Optimum swimming velocity in circular tanks
  = (0.5 to 2.0) x (fish body length)/second



- Velocities in a 'donut-shaped' region about tank center are reduced:
  – allows fish to select a variety of swimming speeds

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Environmental Management: Optimum Swimming Velocity

- Advantages of ideal swimming speed:
  - Increased growth rate & protein synthesis
  - Less aggression
  - Enhanced immune function
  - Improved flesh texture

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Environmental Management: Density

- Increased density, in general
  - Reduced feed intake
  - Reduced FCR
  - Reduced growth
  - Reduced body and fin condition
  - Increased risk of clinical disease outbreaks
- Inconsistent findings in the scientific literature
- For Atlantic salmon, no consistent density effect up to around 80 kg/m^3

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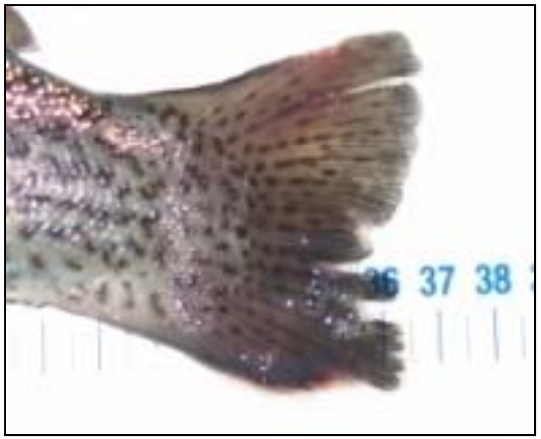
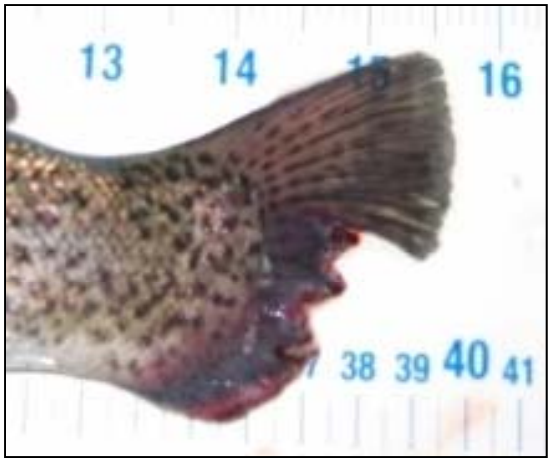
Environmental Management: Density

Fin Erosion

- Common welfare index
- For rainbow trout, optimum densities lie between 40 - 80 kg/m^3
- Need further research on this and other welfare indices for Atlantic salmon





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Environmental Management: Stress Reduction

Types of stress

- **Chemical**
  - Poor diet, suboptimal water quality
- **Biological**
  - Pathogens, aggression, crowding
- **Physical**
  - Light, sound, temperature
- **Husbandry**
  - Handling, transport, treatments

Stress Response:

Depends on the **magnitude** and **duration**

- Stressors are often **cumulative**

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Environmental Management: Stress Reduction

Stress Physiology

- Stressful stimuli result in increased circulating **cortisol** levels
- Elevated cortisol leads to immunosuppresion:
  - Decreased serum bactericidal activity
    - Complement, lysozyme
  - Impaired phagocytosis
  - Decreased white cell count
  - Decreased antibody production

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Environmental Management: Stress Reduction

- Reduce noise & vibration:
  - Use vibration isolation between
    - pumps & pipes
    - motors & floor
    - tank & floor

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Environmental Management: Stress Reduction

- Thoughtful facility design
  - Keep treatment equipment & culture tanks in different rooms
  - Reduce activity around tanks

Courtesy Jim Michaels

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Water Quality
Recirculation Aquaculture

Important Parameters

Dissolved Oxygen

Dissolved Carbon Dioxide

Ammonia / Nitrite / Nitrate

Temperature

pH

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Dissolved Oxygen

- Maintaining optimal DO critical in intensive recirculation systems
- O_2 poorly soluble in water
- Max. DO rarely exceeds 10-12 mg/L in H_2O
- DO consumption rates:
  - Approx. 100 mg O_2/kg body wgt/hour (resting)
  - As high as 800 mg O_2/kg body wgt/hour (actively swimming)
- 85-100% saturation of O_2 required to maintain maximum growth rates in salmonids
- Pure oxygen can be added to system water through low-head oxygenators
  - **Can maintain tank H_2O at saturation**

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Dissolved CO_2

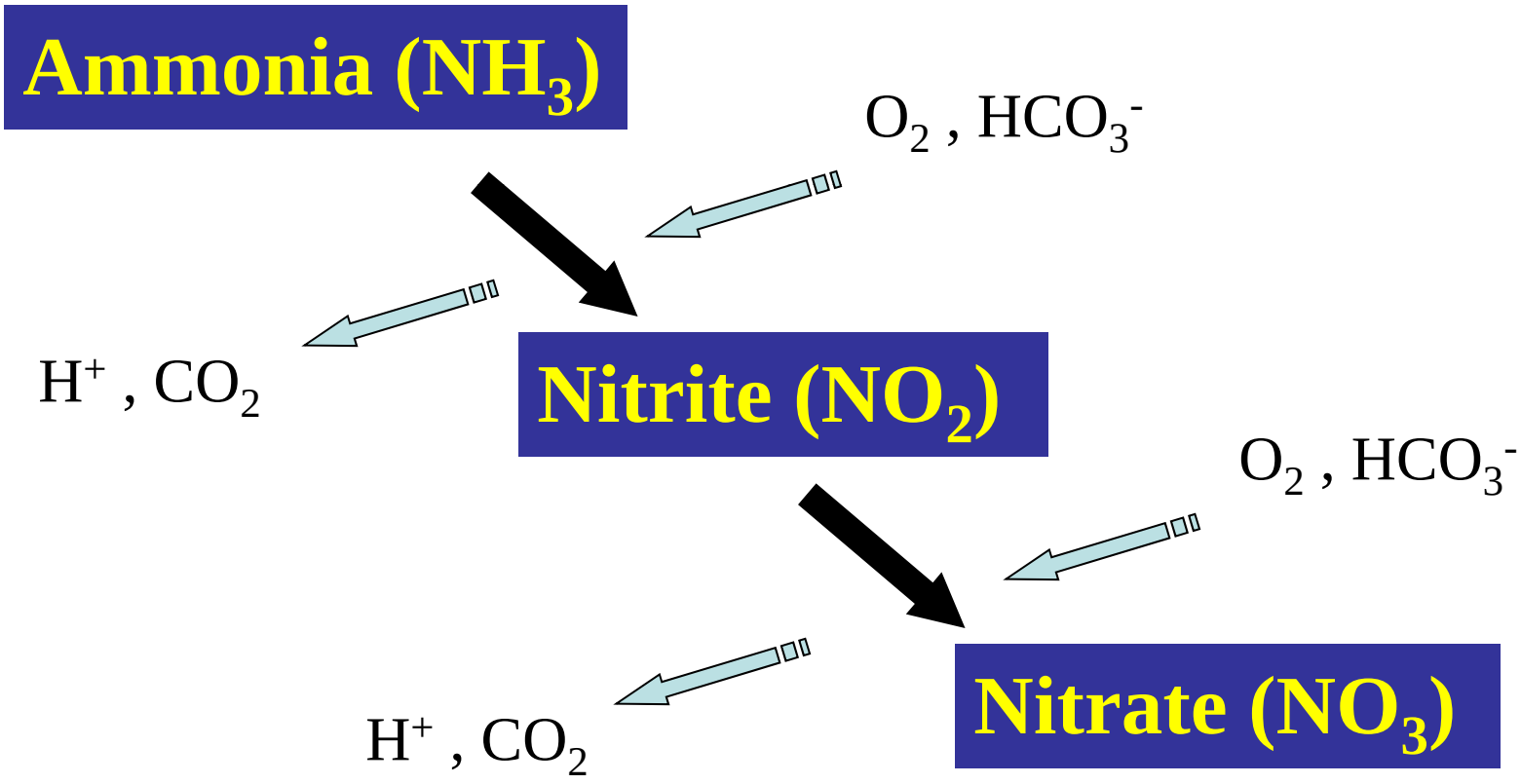
- Salmonids produce 0.96 – 1.10 g CO_2 per gram of O_2 consumed
- Inverse relationship of CO_2 with pH
- Long-term exposure to elevated CO_2
  - Decreased hemoglobin oxygen binding capacity (Bohr effect)
    - Increased ventilation, elevated blood pressure
  - Reduced growth rate
  - Higher FCR
  - Nephrocalcinosis
- Also, increased solubility of toxic metals at lower water pH

Ammonia • Nitrite • Nitrate

$$NH_3 + H^+ + OH^- \leftrightarrow NH_4^+ + OH^-$$

- **Ammonia:** end product of protein catabolism
  - Excreted through gills
  - Rate of nitrogen excretion increases with feeding rate
  - Unionized: NH_3 (most toxic)
    - Direct effects on central nervous system
    - Gill damage
    - Osmoregulatory disruption
  - Ionized: NH_4
  - Balance of unionized vs. ionized: pH dependent

BIOFILTRATION



Ammonia • **Nitrite** • Nitrate

- Nitrite ions taken up through gills, bind to hemoglobin to produce methemoglobin
  - Reduction of blood oxygen transport capacity
  - "Brown blood disease"
- Nitrite toxicity influenced by bicarbonate, potassium, sodium, calcium, and most importantly chloride
  - LC_{50} nitrite: 0.24–12.20 mg/L depending on chloride concentration
  - Chloride cells in gills do not distinguish chloride from nitrite
- Recommended limit: <1 mg/L
  - <0.1 mg/L in soft water

Ammonia • Nitrite • **Nitrate**

- Very low toxicity, and not normally a concern
- Literature: Mortalities occur when levels exceed 1000 mg/L
- Causes non-specific osmoregulatory failure
- Recent evidence at FWI that near-zero exchange RAS (with elevated nitrate) are associated with mortality, deformities, and erratic swimming

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Temperature

- Ectothermic animals
- Increased T = increased metabolism
- Directly effects fish physiological processes:
  - Respiration rate
  - Feed efficiency and assimilation
  - Growth
  - Behavior
  - Reproduction
- Different pathogens thrive at different temperatures

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Temperature

- Water temperature manipulation very important during early life stages of Atlantic salmon:
  - Egg incubation at >8ºC can induce skull and vertebral deformities in fry
  - Increased rearing temperature associated with increased vertebral deformities by 60g in size
    - 12ºC – 2%
    - 14ºC – 13%
    - 16ºC – 15%
    - 18ºC – 22% Baeverfjord et al.

pH

- 6.5 – 8.5 recommended range
- 4.8 > pH > 9.2 can damage and kill salmonids
- pH also has a significant influence on toxicities of e.g. ammonia, hydrogen sulfide, and heavy metals
- In RAS, biofiltration gradually consumes the pH-buffering capacity (i.e. alkalinity) of system water
- Therefore, very important to monitor changes and add sodium bicarbonate when necessary

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Conclusions

- Closed containment recirculation aquaculture shifts biosecurity focus to optimizing environmental conditions
- Further research and system refinements are needed: role of biofilters; accumulating substances (e.g. metals); the effects of alternative feeds of water quality; etc.

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Acknowledgements

All research was supported by the Agriculture Research Service of the United States Department of Agriculture, under Agreement No. 59-1930-5-510.

Opinions, conclusions, and recommendations are of the authors and do not necessarily reflect the view of the USDA.

All experimental protocols involving live animals were in compliance with Animal Welfare Act (9CFR) and have been approved by the Freshwater Institute Animal Care and Use Committee.