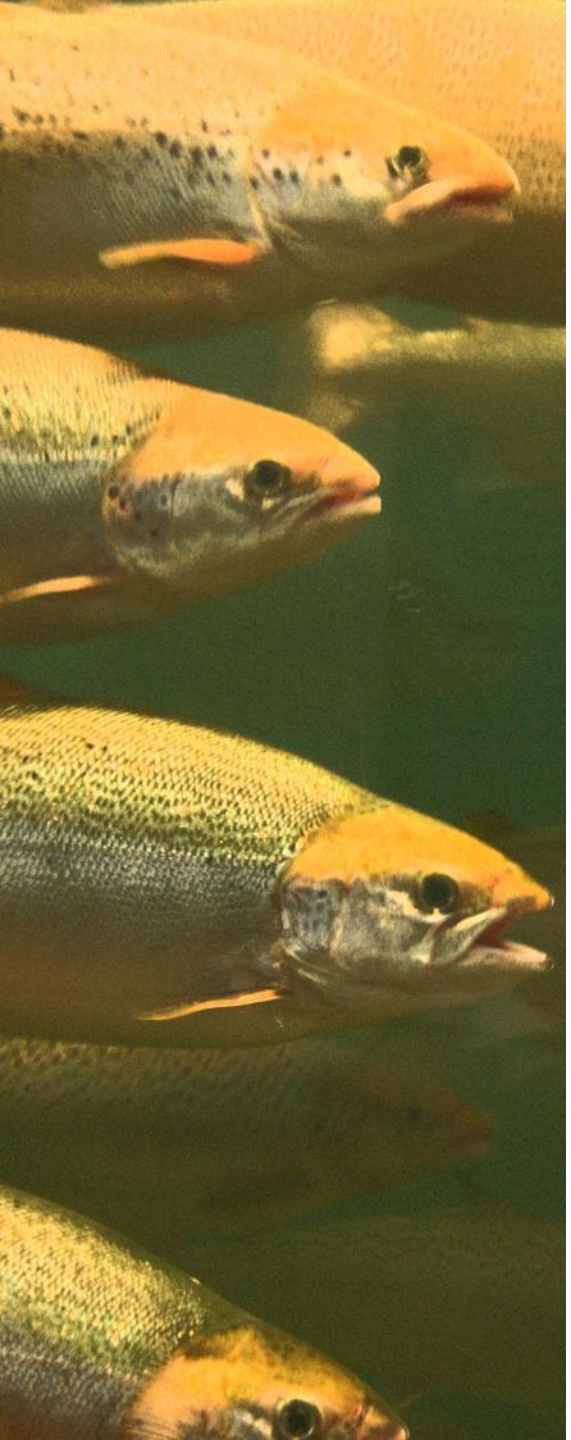


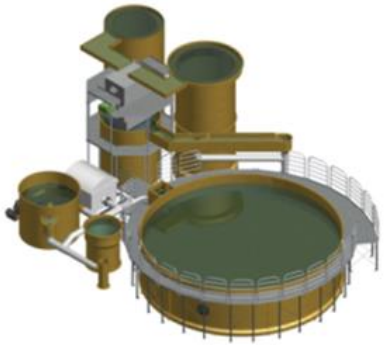
Ozone application in RAS: consequences for fish health and performance

USDA Agricultural Research Service

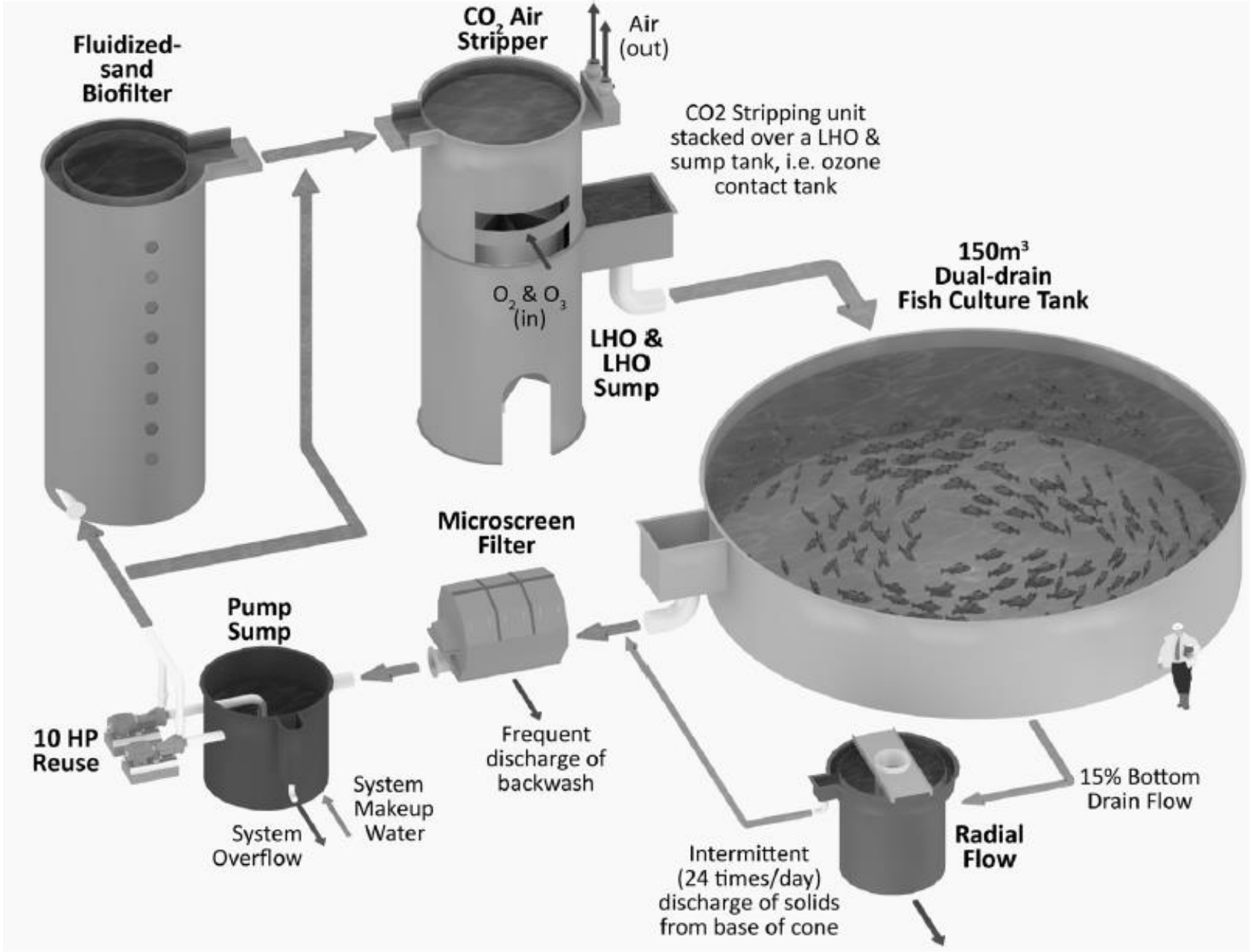
CtrlAQUA
Centre for Closed-Containment Aquaculture

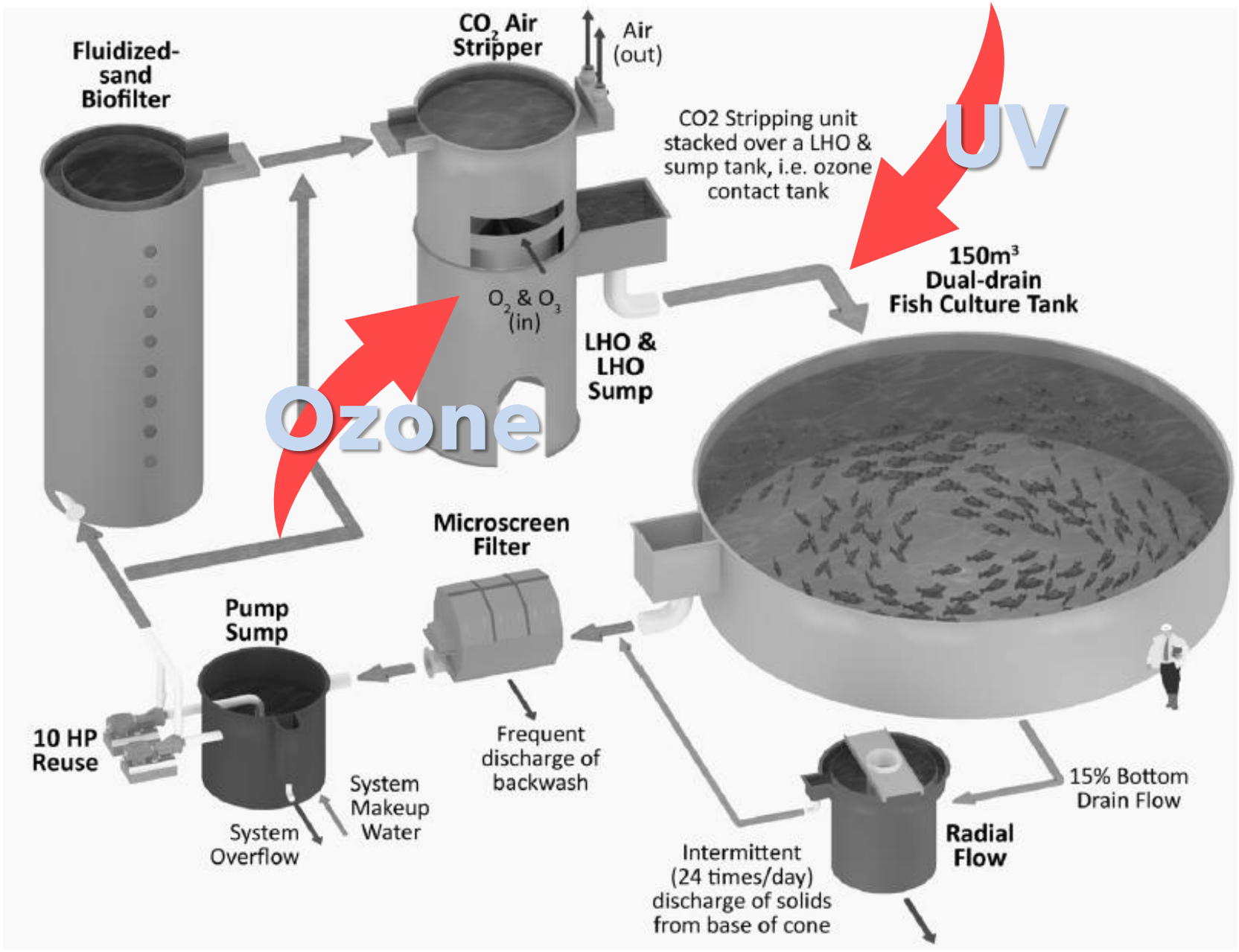
October 17, 2024
Smolt Production Workshop, Sunndalsøra

Chris Good, John Davidson, Steven Summerfelt



The Conservation Fund Freshwater Institute


Fluidized-sand Biofilter
CO_2 Air Stripper
Air (out)
CO2 Stripping unit stacked over a LHO & sump tank, i.e. ozone contact tank
O_2 & O_3 (in)
LHO & LHO Sump
150m³ Dual-drain Fish Culture Tank
Microscreen Filter
Pump Sump
10 HP Reuse
Frequent discharge of backwash
System Makeup Water
System Overflow
15% Bottom Drain Flow
Radial Flow
Intermittent (24 times/day) discharge of solids from base of cone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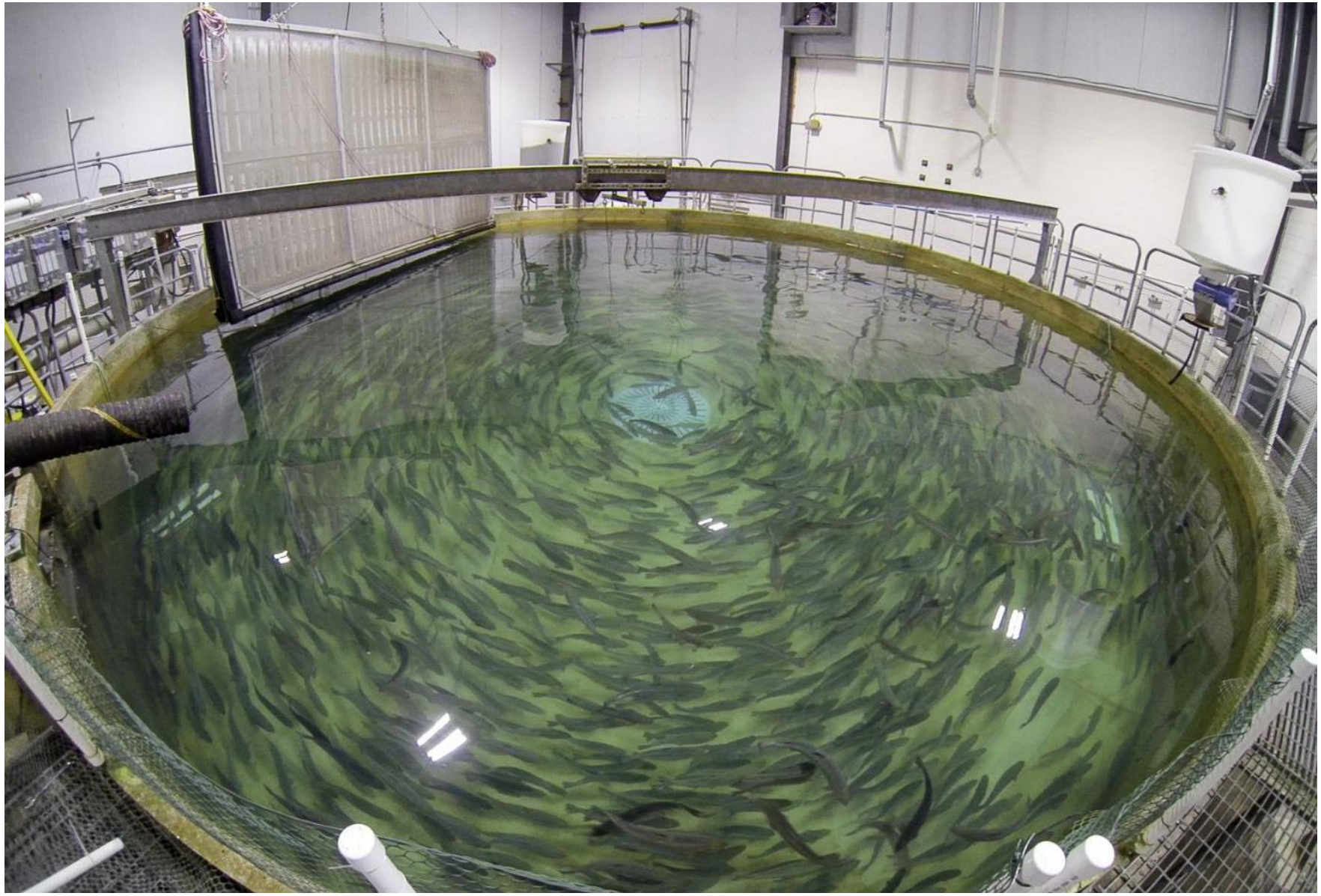


Ozone Improves Water Clarity in RAS with Near-Zero Water Exchange



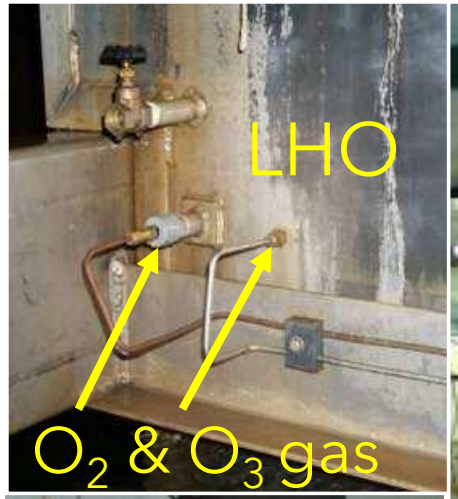
Rapid water clarity progression after turning on ozone in RAS with 90-day system HRT

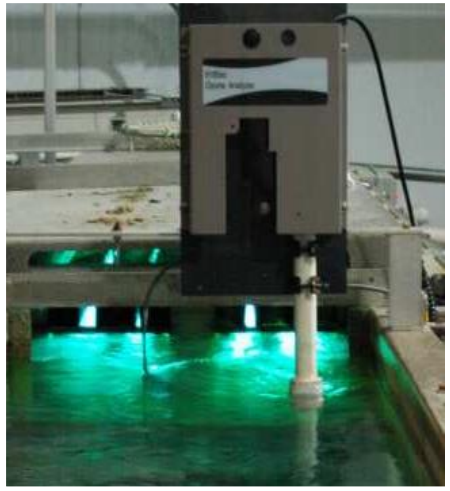

Ozone Advantages

- Rapid reaction rate
  - dissolved ozone half-life = 0–15 sec (Bullock et al., 1997)
- Few harmful reaction by-products in freshwater
- Oxygen produced as reaction end-product
- UV irradiation destroys ozone residuals
- Disinfection capability when combined with UV

Full-Flow O_3 + UV Treatment Study

- Ozone added at LHO alongside O_2 gas
- 1.5 min O_3 contact time in LHO sump
- UV before flow enters fish culture tank to control O_3 residuals
  - ORP control to adjust O_3 generation





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www.elsevier.com/locate/aqua-online

Ozonation followed by ultraviolet irradiation provides effective bacteria inactivation in a freshwater recirculating system

Mark J. Sharrer [1], Steven T. Summerfelt *
The Conservation Fund's Freshwater Institute, 1098 Turner Road, Shepherdstown, WV 25443, United States
Received 23 January 2007; accepted 21 May 2007

• Total Heterotrophic Plate Counts, cfu/ml

	Before Ozone	After Ozone	After UV	% Removal
No Ozone & No UV	466 ± 147	509 ± 139	530 ± 145	NA
Ozone @ 375 mv & No UV	48 ± 9	22 ± 5	21 ± 3	56.3
Ozone @ 375 mv + UV	124 ± 27	81 ± 18	3 ± 1	97.6
Ozone @ 450 mv + UV	50 ± 12	22 ± 4	0 ± 0	100
Ozone @ 525 mv + UV	386 ± 348	225 ±209	0.4 ± 0.3	99.9

Ozone Advantages: Improved Water Quality

- Oxidizes NO_2^- to NO_3^-
- Removes color, fine particles, metals, and dissolved organics:
  - breaks non-biodegradable compounds into smaller, more biodegradable compounds;
  - precipitates dissolved organic molecules;
  - micro-flocculates fine particles;
  - improves solids removal by settling, filtration, or flotation.

Ozone Disadvantages

- Ozone and its reaction by-products can be dangerous to humans and aquatic organisms.
  - Safety measures are required!
- Ozone systems are relatively complicated
- **Toxic byproducts** (brominated compounds) can form when ozonating brackish or saltwater RAS
- Robust materials must be used to deliver ozone
  - Stainless steel, teflon, viton (piping, solenoid valves)

Ozone Disadvantages: Worker Safety

Very low exposure limits

- OSHA 8-h exposure – 0.1 ppm
- 10 min exposure – 0.3 ppm
- Immediately dangerous at 5.0 ppm
  - handheld and in-room ozone gas sensors
  - alarm capabilities and emergency response
  - external shutoffs, ventilation system

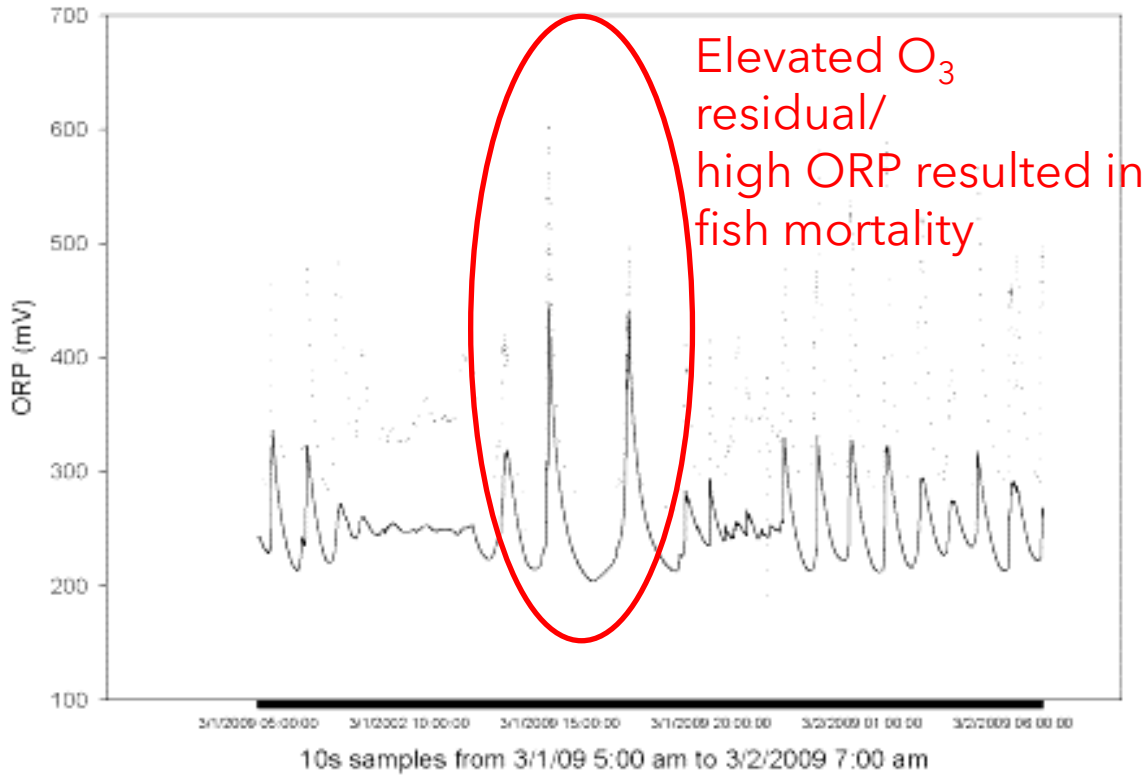
Hand-held ozone sensor

Ozone generator emergency shut-off should be located OUTSIDE the generator room!

Ozone Disadvantages: Toxic to Fish

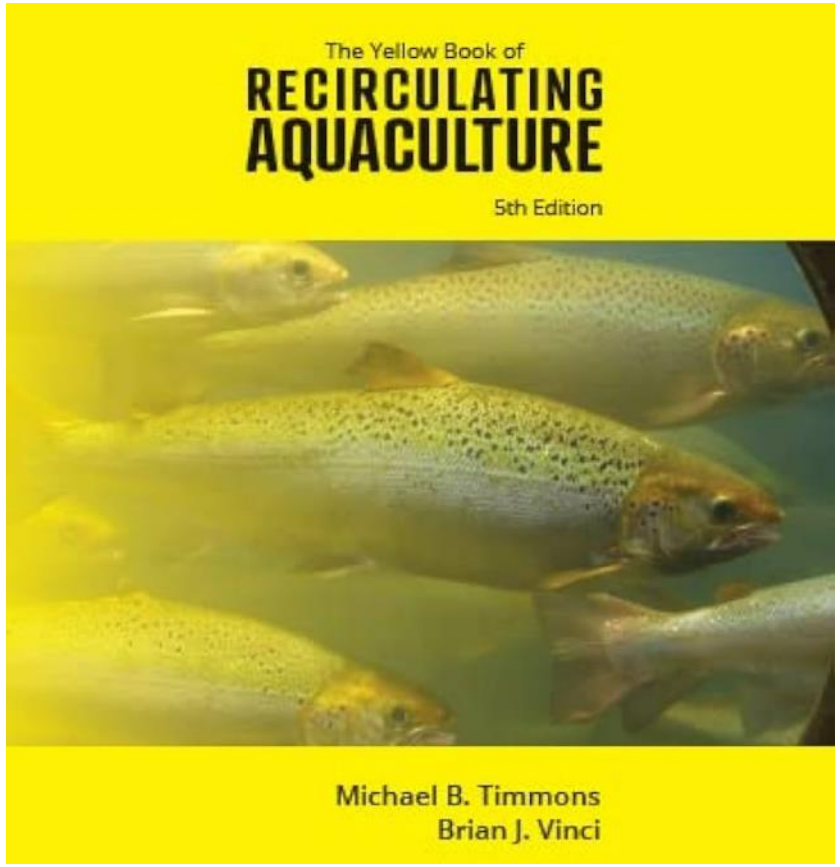
- Toxic to fish at low concentrations
- FI uses ORP (mV) as indirect measure of O_3 residual

Species	Ozone Conc. (mg/L)
Rainbow Trout	0.0093 (96-h LC_{50})
Bluegill	0.06 (24-h LC_{50})
White Perch	0.38 (24-h LC_{50})
Striped Bass Larvae	0.08 (96-h LC_{50})



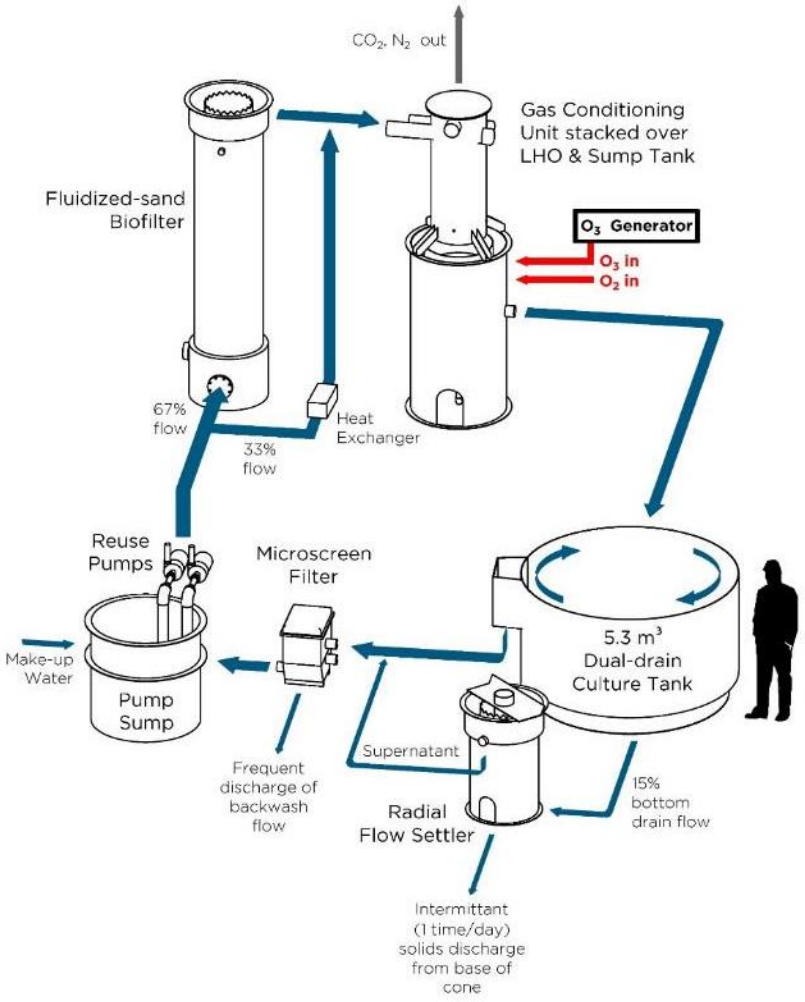
Ozone Dosing Rate

- Summerfelt et al. (2009): 27-29 g ozone/kg feed
  - High dose ozone followed by UV (disinfection)
- Davidson et al. (2011) – 20-25 g ozone/kg feed
  - Low dose ozone, no UV for water quality control
- Yellow Book Rule of Thumb – 13-24 g ozone/kg
  - Low dose for improved water quality and fish health



Ozone Research in Replicate RAS

- 3 RAS with ozone; 3 RAS without ozone
- Non-disinfecting dose with on/off control via ORP feedback
  - Rainbow trout production at various flushing rates
  - Atlantic salmon production
    - Effects on hormones,
    - Early maturation

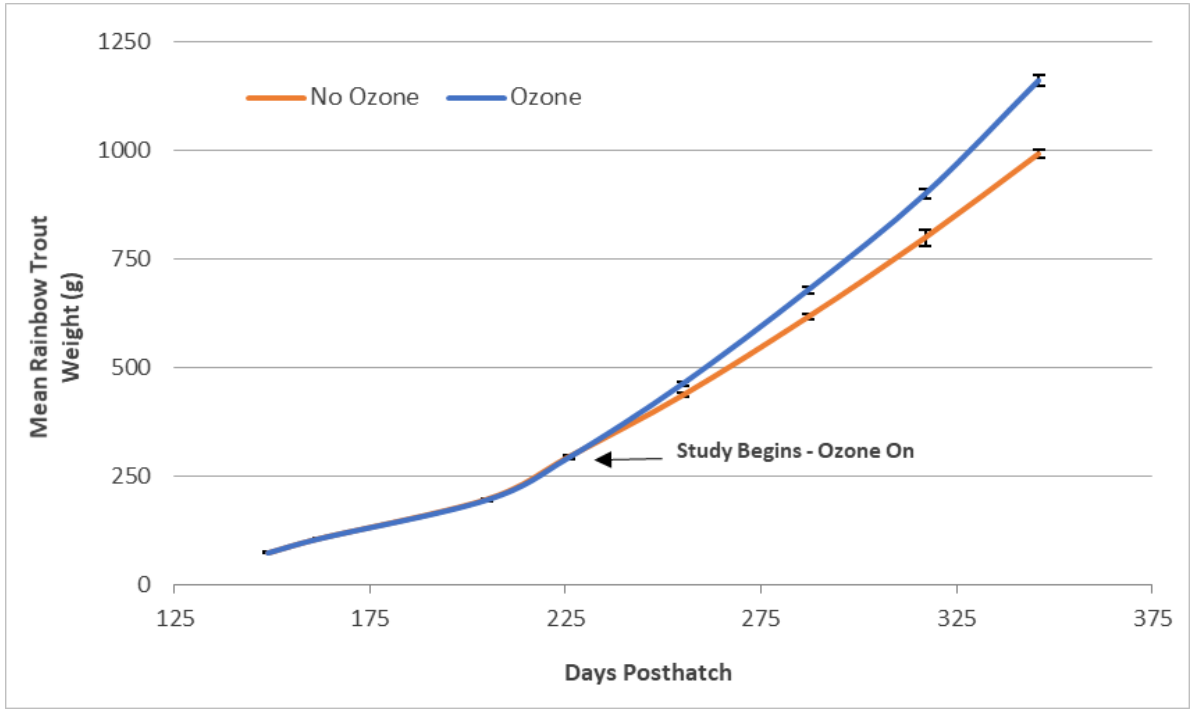


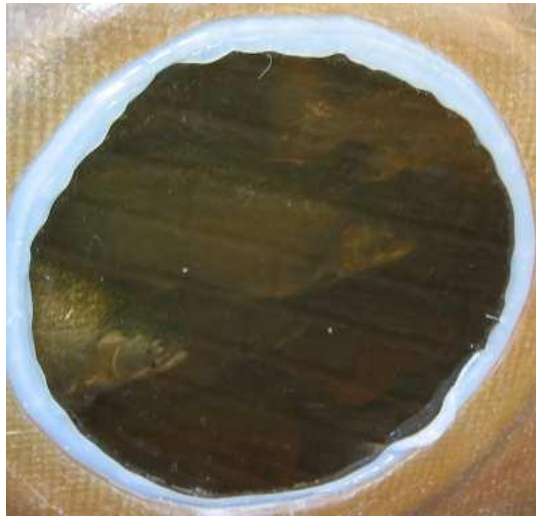
Ozone Improves Water Quality

Davidson et al. (2011). Aqua. Eng. 44, 80-96.	Study 1		Study 2		Study 3	
Water Quality Metrics	Low Exchange No Ozone	Low Exchange Ozone	High Exchange No Ozone	Low Exchange Ozone	Very Low Exchange No Ozone	Near-Zero Exchange Ozone
BOD (mg/L)	3.6	1.7	2.5	3.0	11.8	3.9
True Color (Pt-CO units)	53	4	12	5	157	5
UV Transmittance (%)	60	82	89	77	30	66
TSS (mg/L)	8.7	3.4	3.4	4.6	18.9	3.5
Heterotrophic Bacteria (counts/mL)	2.0×10^5	92	117	114	825	77
Dissolved Copper (mg/L)	0.064	0.021	0.014	0.038	0.119	0.005
Dissolved Zinc (mg/L)	0.005	0.001	0.011	0.007	0.128	0.082

Ozone Improved Rainbow Trout Growth

- Cumulative water quality improvements = faster growth
  - Possibly enhanced feed capture response





Ozone Reduces Waterborne Hormones

- Effects of ozone (*white bars)* vs. no ozone *(gray bars)* on waterborne hormones in RAS water
  - — ozone significantly reduced estradiol levels
  - — testosterone, 11-KT lower in ozonated RAS

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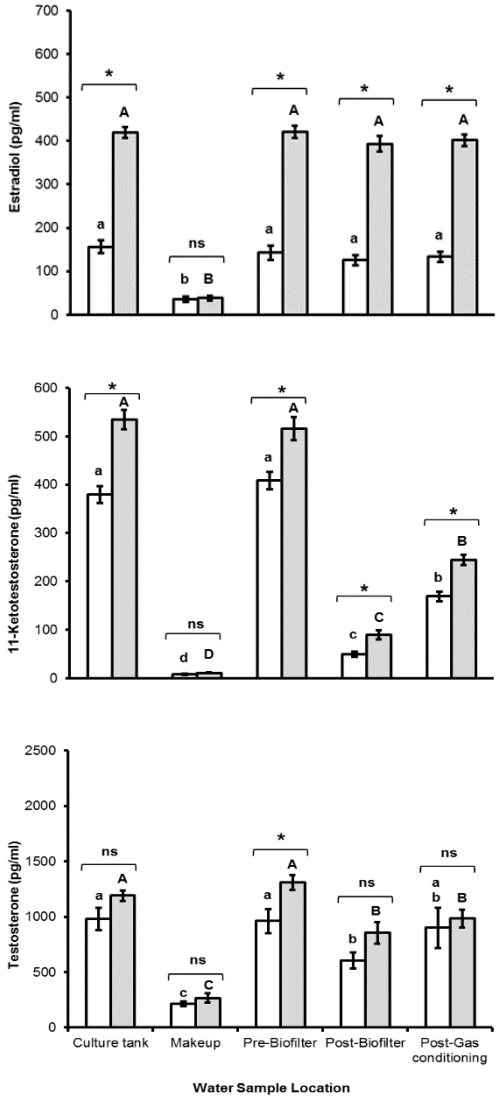
The effects of ozonation on select waterborne steroid hormones in recirculation aquaculture systems containing sexually mature Atlantic salmon *Salmo salar*

Christopher Good[a,*], John Davidson[a], Ryan L. Earley[b], Joseph Styga[b], Steven Summerfelt[a]

[a] *The Conservation Fund's Freshwater Institute, 1098 Turner Road, Shepherdstown, WV 25443, United States*
[b] *Department of Biological Sciences, University of Alabama, 300 Hackberry Lane, Tuscaloosa, AL 35401, United States*

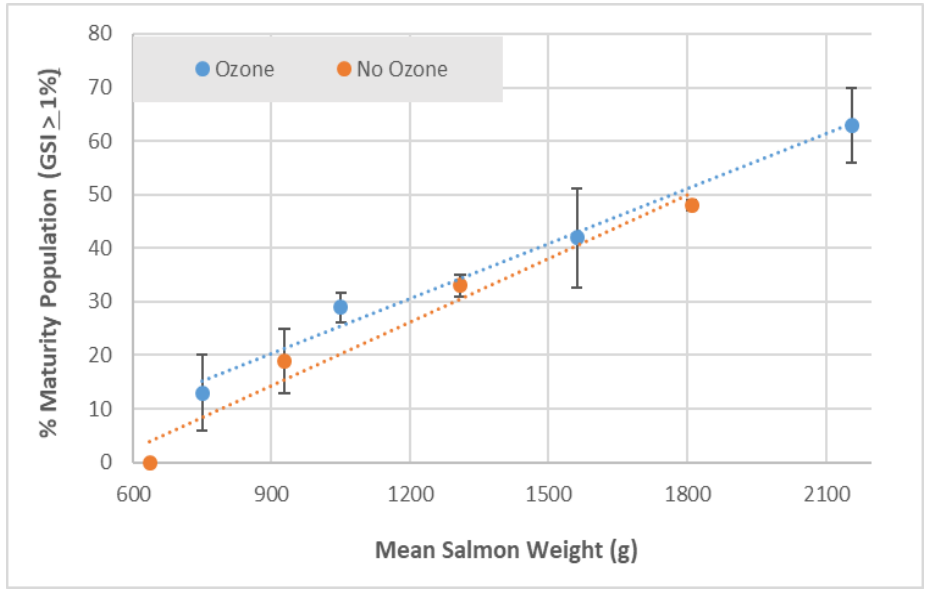


Can Ozone Inhibit Salmon Maturation?

Hormones levels were reduced but early maturation was not inhibited

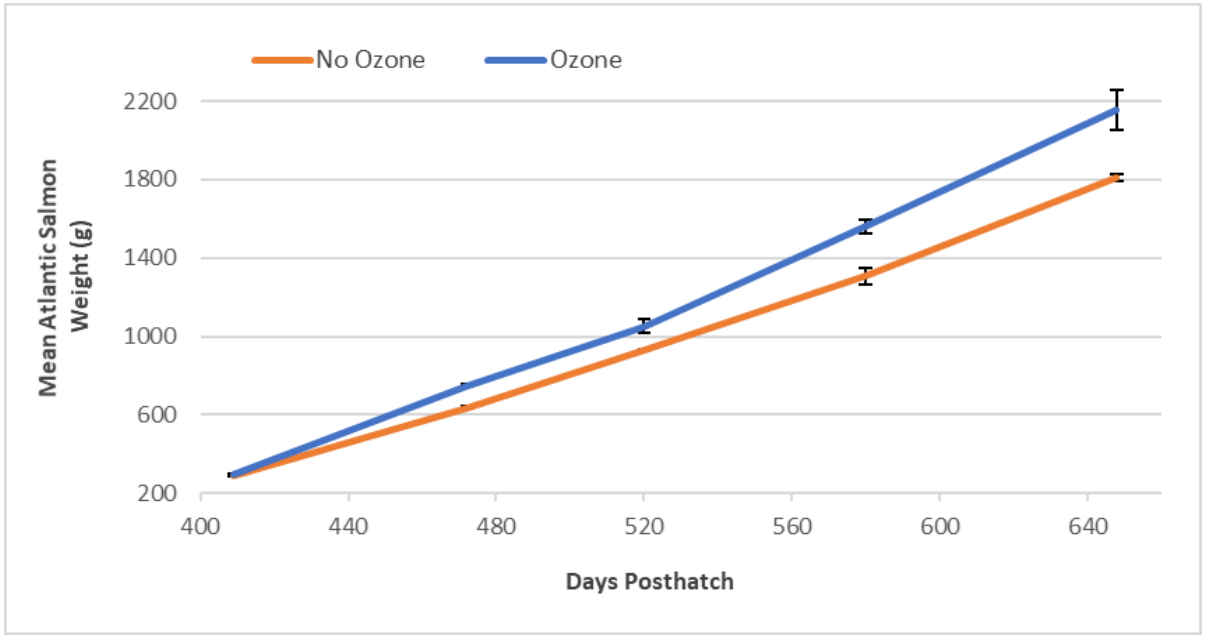
Davidson et al. (2021). Aquaculture 533, 736208.

% Maturity GSI ≥ 1%	2 Months	4 Months	6 Months	8 Months
Ozone	13 ± 7	29 ± 3	42 ± 9	63 ± 7
No Ozone	0 ± 0	19 ± 6	33 ± 2	48 ± 1



Ozone Improved Atlantic Salmon Growth

Water quality was dramatically improved and Atlantic salmon growth was significantly faster in ozonated RAS



Ozone Supports Fish Health & Welfare; however,..

Ozone associated with minor fin erosion

Atlantic salmon (Davidson et al., 2021)

Rainbow trout (Davidson et al., 2011; Good et al., 2011)

Histology also suggests gill changes associated with ozone

		Bimonthly FIn Scores (mean ± standard error)		
Treatment	Welfare Variable	2	4	6
Ozone	Caudal Fin	1.3 ± 0.07	1.5 ± 0.06	1.1 ± 0.23
No Ozone		0.5 ± 0.00	0.8 ± 0.03	0.8 ± 0.00
Ozone	Ventral Fin	1.1 ± 0.06	0.9 ± 0.10	0.8 ± 0.10
No Ozone		0.6 ± 0.03	0.8 ± 0.07	0.7 ± 0.07
Ozone	Left Pelvic Fin	1.3 ± 0.06	1.2 ± 0.06	1.0 ± 0.13
No Ozone		0.9 ± 0.07	1.2 ± 0.09	0.9 ± 0.09
Ozone	Right Pelvic Fin	1.2 ± 0.09	0.9 ± 0.12	0.7 ± 0.12
No Ozone		0.7 ± 0.03	1.1 ± 0.03	0.8 ± 0.15
Ozone	Left Pectoral Fin	0.9 ± 0.10	0.7 ± 0.03	0.6 ± 0.09
No Ozone		0.6 ± 0.07	0.9 ± 0.10	1.0 ± 0.07

THE CONSERVATION FUND

Acknowledgements

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