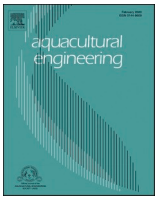
Aquacultural Engineering 95 (2021) 102201

Contents lists available at ScienceDirect

Aquacultural Engineering

journal homepage: www.elsevier.com/locate/aque



Effects of swimming speed and dissolved oxygen on geosmin depuration from market-size Atlantic salmon *Salmo salar*

John Davidson [a,*], Steven Summerfelt [a,1], Casey Grimm [b], Gregory Fischer [c], Christopher Good [a]

[a] The Conservation Fund Freshwater Institute, 1098 Turner Road, Shepherdstown, WV 25443, USA
[b] USDA Agricultural Research Service, Food Processing and Sensory Quality Research Unit, New Orleans, LA 70124, USA
[c] University of Wisconsin Stevens Point Northern Aquaculture Demonstration Facility, 36445 State Hwy 13, Bayfield, WI 54814, USA

ARTICLE INFO

Keywords:
Atlantic salmon
Depuration
Geosmin
Off-flavor
Recirculating aquaculture system
2-methylisoborneol

ABSTRACT

Common off-flavor compounds, including geosmin (GSM) and 2-methylisoborneol (MIB), bioaccumulate in Atlantic salmon *Salmo salar* cultured in recirculating aquaculture systems (RAS) resulting in earthy and musty taints that are unacceptable to consumers. To remediate off-flavor from market-ready salmon, RAS facilities generally relocate fish to separate finishing systems where feed is withheld and makeup water with very low to nondetectable GSM and MIB levels is rapidly exchanged, a process known as depuration. Several procedural aspects that affect salmon metabolism and the associated rate of off-flavor elimination, however, have not been fully evaluated. To this end, a study was carried out to assess the effects of swimming speed and dissolved oxygen (DO) concentration on GSM levels in water and fish flesh during a 10-day depuration period. Atlantic salmon (5–8 kg) originally cultured in a semi-commercial-scale RAS (150 m^3 tank) were exposed to a concentrated GSM bath before being transferred to 12 replicated partial reuse depuration systems (5.4 m^3 total volume). Two swimming speeds (0.3 and 0.6 body lengths/sec) and two DO levels (90% and 100% O_2 saturation) were applied using a 2 × 2 factorial design (N = 3), and each system was operated with a 5-h hydraulic retention time, creating a water flushing to biomass ratio of 151 L/kg fish biomass/day. Geosmin was assessed at Days 0, 3, 6, and 10 in system water and salmon flesh. A borderline effect (P = 0.064; 0.068) of swimming speed was measured for water and fish, respectively, at Day 3, where slightly lower GSM was associated with low swimming speed (0.3 body lengths/sec); however, differences were not detected at Days 6 or 10 when salmon are commonly removed for slaughter. Overall, this research indicates that significant improvements in GSM depuration from RAS-produced Atlantic salmon are not expected when purging with swimming speeds and DO concentrations similar to those tested during this trial.

1. Introduction

Interest in Atlantic salmon *Salmo salar* production using recirculating aquaculture systems (RAS) has increased over the last decade, and several commercial facilities are now producing and selling market-size fish (Summerfelt and Christianson, 2014; Intrafish, 2018). Nevertheless, certain challenges must be overcome to improve the economic viability of this nascent industry, including the tendency for salmon to bioaccumulate unpalatable off-flavors. These earthy and musty taints, which are primarily associated with the microbial metabolites geosmin (GSM) and 2-methylisoborneol (MIB) (Schrader et al., 2005; Schrader and Summerfelt, 2010; Houle et al., 2011; Burr et al., 2012; Lindholm-Lehto and Vielma, 2019), can cause economic loss due to rejected products, negative consumer perception, and increased time, labor, and capital investment for proper remediation (Engle et al., 1995; Tucker, 2000). Presently, the only proven method to eliminate off-flavor from RAS-produced salmon, among other species, is relocation of fish to separate depuration systems where high volumes of water are exchanged while withholding feed (Burr et al., 2012; Davidson et al., 2014; Azaria and van Rijn, 2018; Lindholm-Lehto and Vielma, 2019; Davidson et al., 2020).

Specifically, research evaluating standard operating procedures (SOPs) for Atlantic salmon depuration has established that fish should be purged in separate, recently cleaned (Burr et al., 2012) and disinfected

* Corresponding author.
E-mail address: jdavidson@conservationfund.org (J. Davidson).
[1] Superior Fresh, W15506 Superior Fresh Drive, Hixton, WI 54635, USA.

https://doi.org/10.1016/j.aquaeng.2021.102201
Received 4 June 2021; Received in revised form 23 September 2021; Accepted 29 September 2021
Available online 1 October 2021

systems that are free of high-surface-area media (Davidson et al., 2014) and operated with relatively high water exchange rates (Davidson et al., 2020; Schram et al., 2021). During this process, feed is generally withheld for 6–15 days depending on site- and species-specific variables (Burr et al., 2012; Azaria and van Rijn, 2018; Lindholm-Lehto and Vielma, 2019; Davidson et al., 2020); however, extended feed deprivation can result in fish weight loss (Burr et al., 2012; May, 2020), reduced lipid levels in fillets (Burr et al., 2012), and increased water and energy use (Davidson et al., 2020), all of which have economic consequences for RAS operations. As such, procedural refinements that reduce the required duration of Atlantic salmon purging would be highly beneficial.

Given that fish primarily uptake and eliminate lipophilic chemicals, including GSM and MIB, across their gills (Randall et al., 1998; Streit, 1998; Howgate, 2004), it is important to evaluate conditions that influence gill ventilation rate including: (i) increased swimming speed induced by water velocity, and (ii) dissolved oxygen (DO) level of the culture environment. Anecdotal evidence of a positive effect of swimming speed on off-flavor in adult Atlantic salmon (2–3 kg) was noted during a study evaluating the effects of 0.40–0.45 vs. < 0.1 body lengths/sec on salmon growth and muscle composition. Totland et al. (1987) reported that taste panelists detected less off-flavor in exercised fish; albeit, the analysis was non-descriptive, i.e., based on eating preference, and GSM and MIB concentrations were not measured. Further, Schram et al. (2016) evaluated the effects of exercise while depurating European eel *Anguilla anguilla* and found that increased swimming speed enhanced the rate of GSM elimination. Schram et al. (2016) also noted that exercise increased oxygen consumption rate, which was linearly correlated with GSM excretion. Moreover, many studies have demonstrated that branchial transfer of lipophilic chemicals in fish is related to oxygen consumption rates (e.g., Randall et al., 1998; Yang et al., 2000); however, gill physiology, ventilation rate, and the associated transfer of lipophilic compounds can vary among species (Schultz and Hayton, 1999). Interspecies differences in depuration dynamics are also expected relative to fillet lipid content (Gobas and MacKay, 1987; Johnsen and Lloyd, 1992; Howgate, 2004) and water exchange rate (Schram et al., 2016; Davidson et al., 2020).

Within this framework, a research trial was carried out to evaluate the effects of swimming speed and DO on GSM depuration from market-size Atlantic salmon originally produced in a semi-commercial-scale RAS. The authors expected this work to guide optimization of depuration SOPs for RAS-produced Atlantic salmon and hypothesized that increased swimming speed and lower oxygen levels would facilitate a shorter depuration period.

2. Materials and methods

2.1. Experimental systems and design

Prior to the study, 12 replicated partial reuse systems (PRAS) (Fig. 1) were cleaned and disinfected by recycling water with 250 mg/L hydrogen peroxide per procedures suggested by Davidson et al. (2014). Two swimming speeds dictated by water rotational velocity (0.3 and 0.6 body lengths/sec) and two DO levels (90% and 100% O_2 saturation) were applied using a 2 × 2 factorial design (N = 3). Six PRAS were equipped with submersible velocity boosting pumps (Model SP40A2 20–01, Pentair Hydromatic®, Pentair Aquatic Ecosystems, Apopka, FL, USA) located in the tank side-box beside the water recycle pump (Fig. 1). A 5.1 cm dia. outlet pipe with 13 equally spaced orifices (1.3 cm dia.) was positioned parallel to the tank wall. This pumping system doubled the tank's rotational velocity to 45 ± 1 cm/sec compared to normally operated PRAS (24 ± 1 cm/sec) per measurements collected at the beginning and end of the study using a Pygmy Meter with Model 100 Flow Indicator (Gurley Precision Instruments, Troy, NY, USA) positioned parallel to the tank wall (15 cm from the side and 30 cm deep). Fish swimming speed was calculated as:

Fish Length (cm)/ Water Rotational Velocity (cm/sec) = Body Lengths/sec (bls).

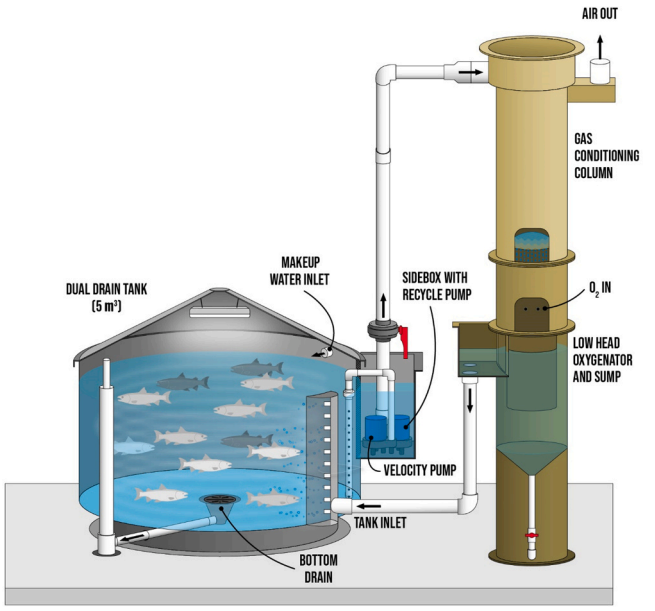


Fig. 1. Water flow, process design, and velocity boosting infrastructure for an individual partial reuse depuration system.

Partial reuse systems within each velocity treatment were operated with 90 or 100% O_2 saturation, respectively, via adjustment of oxygen gas flow to the low head oxygenator (LHO), where gas flow was turned off to create 90% O_2 saturation. Dissolved oxygen and water temperature were recorded daily from an LC3 Central Monitoring System integrated with a Point Four™ RIU3 (Pentair Aquatic Ecosystems, Apopka, FL, USA) and a PRO-X DO probe (In Situ, Fort Collins, CO, USA). Additionally, PRAS recirculation and makeup water flows were measured and calibrated using a Model DXN Portable Ultrasonic Flow Meter (Dynasonics, Racine, WI, USA). Each PRAS recirculated 340 ± 5 L/min, and average makeup water addition was 18.6 L/min (0.0186 m^3/min), resulting in a 5-h hydraulic retention time through the 5.4 m^3 system and a water flushing to biomass ratio of 151 L/kg fish biomass/day.

2.2. Atlantic salmon

All-female Atlantic salmon (Stofnfiskur, Hafnarfjörður, Iceland) were received as fertilized eggs, hatched onsite in a chilled water incubation system, and then cultured to market-size in freshwater systems. To begin the study, 312 fish (5–8 kg; 6.8 kg mean weight) were harvested from an indoor 260 m^3 semi-commercial scale RAS (Davidson et al., 2016) where they had been fed a commercial 44% protein/ 29% fat diet (EWOS Dynamic Red; Cargill Inc., Minneapolis, MN, USA). The fish were transported to an 18 m^3 tank within a partial reuse system. One day later, salmon were exposed to a concentrated GSM bath to ensure that initial flesh levels were representative of fish requiring depuration. Water flushing was discontinued for four hours and a concentrated GSM solution (4 ml of 2 mg/ml GSM; Sigma-Aldrich, St. Louis, MO, USA) was added. Similar procedures to bioconcentrate GSM in fish flesh have been described (Schram et al., 2017; Davidson et al., 2020). Immediately following GSM exposure, salmon were randomly stocked (26 fish per tank; ~177 kg total biomass) among 12 replicated PRAS (Fig. 1) at a density of 33 kg/m^3/tank. A 10-day off-feed depuration period followed.

2.3. Fillet off-flavor and proximate compositional analysis

Immediately following GSM exposure, six fish were removed,

humanely euthanized, and processed to obtain flesh samples for baseline (Day 0) GSM and MIB assessment. Thereafter, three fish per PRAS were collected and processed on Days 3, 6 and 10 post-stocking using the same procedures. All sampled fish were randomly selected; however, only fish that displayed morphometric traits common to immature, premium quality fish (Aksnes et al., 1986) were kept for analysis. Using the pectoral fin as a reference point, a consistent anterior fillet portion (Davidson et al., 2020) was removed, vacuum sealed, and frozen in preparation for shipment to the Food Processing and Sensory Quality Research Lab (United States Department of Agriculture, Agriculture Research Service, New Orleans, LA, USA). A modified method of Lloyd and Grimm (1999) following solid phase microextraction, gas chromatography, mass spectrometry (SPME/GC/MS) procedures as described in Davidson et al. (2020) was used to quantify off-flavor in salmon fillets. Limits of detection and quantification for GSM and MIB in fillets were 1 and 5 ng/kg, respectively. In addition, on Day 10, a consistent skin-on fillet section guided by dorsal and pectoral fin insertion points was collected from the same fish and sent to West Virginia University's Division of Food Sciences for proximate compositional analysis of crude fat using methods described in Association of Analytical Chemists (1990).

2.4. Off-flavor sampling and analysis - water

Glass scintillation vials (20 ml) were used to collect water for off-flavor analysis. Water samples were collected before dosing GSM concentrate (Time 0) and every 30 min thereafter until the 4-h exposure period was complete (Fig. 2). During the experiment, water samples were collected from a common location in each PRAS tank before stocking fish (Day 0) and thereafter on Days 3, 6, and 10. Parafilm was wrapped around the lid to prevent leakage and contamination, and vials were refrigerated (~4 °C) prior to shipment for analysis. Samples were tested using SPME/GC/MS (Lloyd et al., 1998), as described in Davidson et al. (2020). Limits of GSM and MIB detection and quantitation in water were 1 and 2 ng/L, respectively.

2.5. Background water quality

Water samples were also collected from PRAS side drains and makeup water on Days 2 and 9 and tested for carbon dioxide (CO_2), total alkalinity (ALK), total ammonia nitrogen (TAN), total phosphorus (TP), and total suspended solids (TSS) to characterize the depuration system environment so that commercial salmon RAS operations can effectively compare conditions. Standard procedures described by American Public Health Association (2012) and HACH Company (2003, 2015) were followed for these analyses.

2.6. Statistical analysis and off-flavor decay modeling

Off-flavor concentrations in fish flesh and water, background water quality metrics, and crude fat levels were averaged per PRAS (tank) at each sampling point, and a grand mean and standard error were calculated for each treatment (N = 3). A balanced two-way analysis of variance test was applied to each data set using SYSTAT 13 software (2009), and a probability level of 0.05 was used to determine significance. In addition, the natural log of fillet geosmin levels at each sampling point was calculated per replicate PRAS and linear regression analysis was carried out to produce respective off-flavor decay slope coefficients and associated constants. Slope coefficients were averaged by treatment and analyzed with a 2-way ANOVA (N = 3), and mean regression curves were modeled using the following equation:

$$[A] = [A]_o \exp(-kt),$$

where [A] is the predicted concentration of geosmin in the salmon flesh on the y-axis, $[A]_o$ is the starting GSM concentration in fish flesh dictated by the regression constant, k is the mean decay slope coefficient from the regressions, and t is time in days on the x-axis. A similar modeling approach was carried out by adjusting the initial GSM starting concentration, while maintaining the other factors for the 0.3 bls, 100% O_2 saturation treatment.

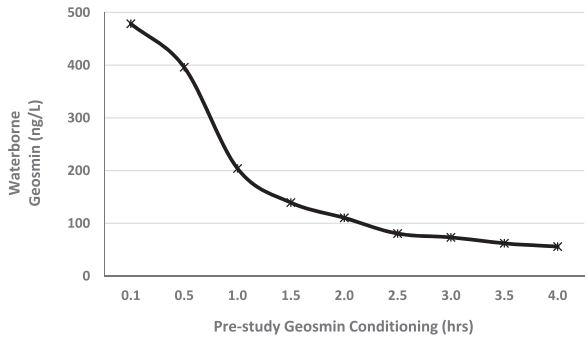


Fig. 2. Waterborne GSM concentration (ng/L) expressed with time during the pre-study exposure period utilized to boost GSM levels in salmon flesh.

3. Results

3.1. Geosmin - water

During the 4-h off-flavor exposure period, salmon rapidly absorbed waterborne GSM, as evidenced by declining tank water concentrations (Fig. 2). After fish were stocked into the respective depuration systems, waterborne GSM initially increased (Day 3) and declined thereafter (Fig. 3). Differences in GSM were not detected in tank water relative to fish swimming speed or DO concentration, and interactive effects of these factors were not observed. Nevertheless, a borderline effect of swimming speed was measured at Day 3 ($P = 0.064$) where depuration systems operated with lower swimming speeds (0.3 bls) reflected slightly lower GSM levels in tank water. Only trace levels of MIB were periodically detected in water samples; therefore, MIB levels were not reported.

3.2. Geosmin – salmon

The off-flavor exposure procedure effectively boosted GSM levels above the sparsely reported and wide-ranging recommended taste threshold for salmonids, i.e., 250–900 ng/kg (Robertson et al., 2005; Petersen et al., 2011; Burr et al., 2012). The initial GSM concentration in Atlantic salmon flesh at Day 0 was 1208 ± 150 ng/kg (Fig. 4). During the experiment, differences in GSM were not detected in salmon flesh relative to swimming speed or DO concentration, and interactive effects

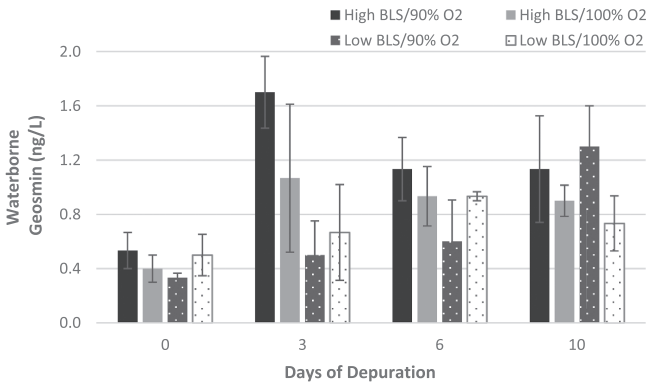


Fig. 3. Geosmin concentrations (ng/L; mean ± standard error) in depuration system water over the 10-day depuration period in PRAS operated with different combinations of salmon swimming speed and dissolved oxygen concentrations.

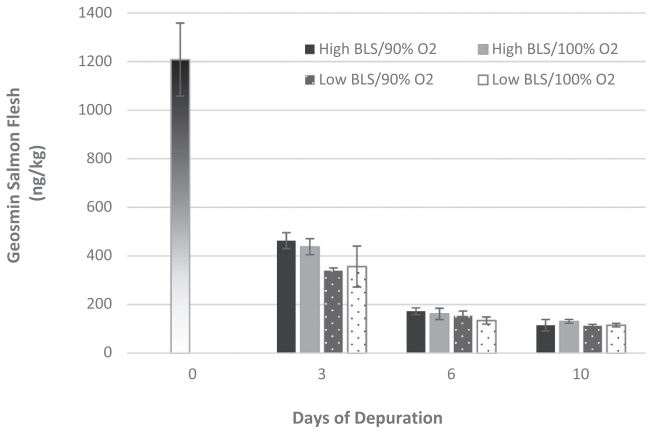


Fig. 4. Geosmin levels (ng/kg; mean ± standard error) in salmon flesh over the 10-day depuration period in PRAS operated with different combinations of fish swimming speed and dissolved oxygen concentrations.

of these factors were not observed. However, a borderline effect of swimming speed was measured in fish flesh at Day 3 ($P = 0.068$), matching the trend reported for water. Further, linear regression analysis indicated that Atlantic salmon weight within the tested range did not correlate with GSM level in fish flesh at Days 3, 6, and 10 where R^2 values were 0.047, 0.010, and 0.040, respectively. Overall, salmon subjected to each swimming speed *x* DO combination effectively purged GSM from their flesh following typical trends for exponential off-flavor decay (Howgate, 2004) (Fig. 5). Only trace levels of MIB were periodically detected in fish flesh; therefore, MIB levels were not reported. In addition, lipid levels in Atlantic salmon flesh were not affected by swimming speed or DO. Average fillet lipid level measured after ten days of depuration was 16.6 ± 0.5%.

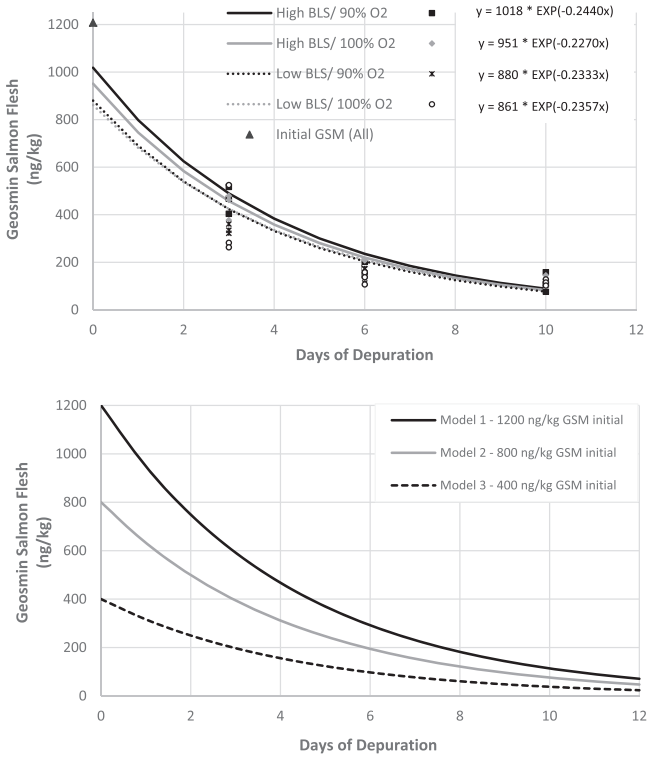


Fig. 5. Predicted exponential off-flavor decay in salmon flesh for each swimming speed x dissolved oxygen treatment (top) and modeled off-flavor decay for the 0.3 body length/sec x 100% O_2 saturation treatment at various initial geosmin levels in fish flesh (bottom).

3.3. General water quality

Background water quality was generally similar between treatments. For example, no significant effects of swimming speed or DO were detected for CO_2, ALK, and TP; however, higher TAN and water temperature were measured in depuration systems with faster water velocity and swimming speed. Significantly higher TSS was measured in PRAS operated with 100% O_2 saturation (Table 1). It is important to note that the magnitude of differences in background water quality was relatively low, i.e., ~0.05 mg/L TAN, ~0.6 °C, and < 1 mg/L TSS. No interactive effects of swimming speed and DO were found when analyzing these metrics.

4. Discussion

Overall, swimming speed and DO did not affect the rate of geosmin elimination by Atlantic salmon during this study. These findings contradict our hypothesis that adjustment of variables that increase fish metabolism and gill ventilation rate would enhance geosmin depuration from market-size salmon. Further, Day 3 data suggest that lower swimming speed (0.3 bls) nearly reduced GSM in water and fish, as evidenced by *P*-values slightly above the specified significance level. It is worth noting, however, that PRAS operated with 0.3 bls and 90% O_2 saturation had the lowest residual off-flavor in tank water (Fig. 3) before fish were stocked (Day 0), and that trend reflected similarly at Day 3 (Fig. 4). As such, lower background GSM levels associated with this treatment may have dictated the borderline statistical effect. Nevertheless, this trend was not repeated at Days 6 and 10 which are more representative of typical salmon harvest times.

Further, statistical analysis of off-flavor decay slope coefficients generated via regression analysis validated that there was no effect of swimming speed or DO on the rate of GSM decay in fish flesh (Table 2). A similar exercise was carried out to model GSM decay for the 0.3 bls, 100% O_2 saturation treatment using the respective slope coefficient (−0.236) at varying initial GSM levels in salmon flesh, i.e., 1200, 800, and 400 ng/kg, respectively (Fig. 5), where Model 3 represents similar depuration SOPs and approximate starting GSM level (400 ng/kg) in salmon at the Freshwater Institute (Fig. 5). Interestingly, Fig. 5 (bottom) indicates that salmon would likely be depurated by Day 6 under these conditions (assuming that 100 ng/kg is below human detection), which aligns with onsite experience and establishment of a 6-day onsite purge cycle.

The reason that we did not observe improvements in geosmin elimination, however, remains unknown. In theory, conditions of increased swimming speed and lower DO should have increased fish metabolism and gill ventilation rate. As mentioned, Schram et al. (2016) found that European eel exercised at optimal swimming speeds excreted geosmin at significantly faster rates compared to eel that were kept at near-static velocities. Several important differences exist between Schram et al. (2016) and the present study, however. First, there are obvious dissimilarities between Atlantic salmon and European eel relative to body size, physiology, morphology, and fat content. Schram et al. (2016) also suggested that European eel metabolize or biotransform geosmin, while excretion appears to be the primary mechanism of elimination in Atlantic salmon (Davidson et al., 2020; Schram et al., 2021). Further, Schram et al. (2016) subjected eels to swimming speeds that were approximately 11 times different, whereas the present study utilized a two-fold difference, which may not have been enough to influence salmon metabolism and gill ventilation rate. Unfortunately, the authors found it difficult to effectively measure gill ventilation rate without disrupting normal swimming activity while standing near the tanks.

The relatively small difference in swimming speed that was assessed during this study was primarily related to feasible depuration system operation with large, market-size salmon. A maximum swimming speed of 0.6 bls was achieved only when utilizing an additional pump capable of moving approximately 350 L/min of water. Moreover, Atlantic

Table 1
Background water quality concentrations (mean ± standard error; N = 3) measured in depurations systems for each flushing/ HRT treatment.

Swimming Speed (body lengths/sec)		0.6	0.6	0.3	0.3	-
Dissolved Oxygen (% Sat. O_2)		90	100	90	100	Makeup Water
Carbon Dioxide (mg/L)		5.7 ± 0.4	5.0 ± 0.8	5.8 ± 0.9	5.7 ± 0.7	17.3
Dissolved Oxygen (mg/L)	b	9.0 ± 0.10	10.0 ± 0.07	9.2 ± 0.02	10.3 ± 0.04	–
Total Alkalinity (mg/L)		249 ± 3	250 ± 4	247 ± 3	243 ± 2	246
Total Ammonia Nitrogen (mg/L)	a	0.33 ± 0.04	0.32 ± 0.01	0.31 ± 0.02	0.24 ± 0.01	0.08
Total Phosphorous (mg/L)		0.022 ± 0.004	0.030 ± 0.004	0.019 ± 0.004	0.024 ± 0.003	0.008
Total Suspended Solids (mg/L)	b	1.3 ± 0.2	2.6 ± 0.4	1.8 ± 0.2	2.2 ± 0.4	1.3
Water Temperature (° C)	a	15.3 ± 0.06	15.2 ± 0.02	14.7 ± 0.01	14.6 ± 0.11	13.6

Superscripts indicate parameters significantly impacted by swimming speed (a) or dissolved oxygen (b). Lack of superscripts indicates no significant treatment effects.

Table 2
First order decay slope coefficients calculated for each swimming speed x DO treatment.

Swimming speed (body lengths/sec)	Dissolved oxygen (% Sat. O_2)	First order decay slope coefficient
0.6	90	-0.244 ± 0.020
0.6	100	-0.228 ± 0.004
0.3	90	-0.233 ± 0.009
0.3	100	-0.236 ± 0.012

Table 3
Approximate water rotational velocities required to achieve 0.5, 1.0, and 1.5 bls for various sized market-ready Atlantic salmon, based on authors' personal data.

Atlantic Salmon Weight (kg)	3	4	5	6	7
Approximate Salmon Length (cm)	56	63	67	71	76
Velocity (cm/sec) to Achieve 0.5 bls	28.0	31.5	33.5	35.5	38.0
Velocity (cm/sec) to Achieve 1.0 bls	56.0	63.0	67.0	71.0	76.0
Velocity (cm/sec) to Achieve 1.5 bls	84.0	94.5	100.5	106.5	114.0

salmon used for this trial were within the range of commercial harvest-size, but slightly larger (5–8 kg) and longer (~ 75 cm) than average market-size salmon (4–5 kg) (Mowi, 2020; NASDAQ, 2021), which are generally 63–67 cm long (Table 3). Nevertheless, regression analysis indicated that salmon size (weight) did not correlate with GSM levels in the flesh, and swimming speeds would only have increased slightly to 0.4 and 0.7 bls if 4 kg salmon were used for the study (Table 3). Recommended swimming speeds for farmed Atlantic salmon of various life stages have been reported from 0.75 to 2.0 bls relative to improvements in growth, health, welfare, and precocious maturation (Jobling et al., 1993; Solstorm et al., 2015; Waldrop et al., 2018; Timmerhaus et al., 2021), and Davison (1997) concluded that exercise training up to 1.5 bls normally conveys benefits for fish. For practical purposes, if a salmon RAS producer wished to achieve a swimming speed of 1 bls, which Timmerhaus et al. (2021) suggested as a possible optimum, significant adjustments to depuration system design and/or operation may be required to increase rotational velocity. Considering the system design employed during this study, further increases in rotational velocity could be achieved via reduced nozzle orifice size to increase water jet velocity at the tank inlet, elevated LHO sump height and capacity to increase water head pressure above the tank, and larger sizing of water recycle pumps to raise system flow rates. Alternatively, a larger water velocity boosting pump could be utilized to increase target velocities without impacting the primary system design. It is important to note that use of velocity boosting pumps during the present study imparted heat transfer that increased water temperature by 0.6 °C (Table 1), but this did not affect the rate of geosmin elimination.

Similar to the difference in velocity, the DO concentration gradient applied during this trial was also relatively low. The original study design proposed use of 70% O_2 saturation; however, the authors discovered that this was unachievable due to available number of salmon, associated biomass, and reduced oxygen demand of fish that were not feeding. While lower DO levels could be achieved in fully stocked depuration systems at biomass densities of 80–100 kg/m^3, producers would need to consider the risk of maintaining market-ready fish at reduced oxygen levels, as pump or other system failures could result in catastrophic loss of fish.

Although significant effects of the tested swimming speeds and DO levels on GSM remediation were not observed during this study, additional research may be warranted to evaluate the effect of these variables with larger treatment gradients. For example, future research could evaluate swimming speeds of 1.0 vs. 0.3 bls and DO levels of 70–80 vs. 100% saturation. Improvements to depuration system design or size of the velocity boosting pump would be required, as well as increased fish numbers and biomass to support further reduction of oxygen levels. Costs of infrastructure enhancement and associated energy use related to larger recycle or velocity boosting pumps would need to be weighed against possible improvements in off-flavor remediation. Future research could also consider the use of heart rate monitors or non-invasive video capture techniques to assess metabolic and gill ventilation rates, respectively.

5. Conclusions

Significant effects of the tested variables were not observed during this study; therefore, improvements in geosmin elimination rate are not expected when maintaining fish swimming speeds of 0.3–0.6 bls and oxygen levels of 90 or 100% saturation. While swimming speed could be increased slightly and oxygen level reduced, practical gains related to adjustment of these criteria are unlikely due to large fish size (length), economic tradeoffs related to increased capital and energy investments, and associated risk of maintaining lower oxygen levels for market-ready fish. Overall, these findings indicate that adjustments to swimming speed and dissolved oxygen levels within the tested range will not shorten the depuration cycle for RAS-produced Atlantic salmon.

CRediT authorship contribution statement

John Davidson: Methodology, Formal analysis, Investigation, Writing – visualization, original draft, & editing, **Casey Grimm**: Investigation, Resources, Writing – review & editing. **Steven Summerfelt**: Conceptualization, Methodology, Writing – review & editing. **Gregory Fischer**: Funding acquisition, Methodology, Writing – review & editing. **Christopher Good**: Project administration, Supervision, Conceptualization, Writing – review & editing.

Declaration of Competing Interest

The authors declare that there are no conflicts of interest to report.

Acknowledgments

Special thanks to Natalie Redman, Megan Murray, and Destiny Evy for water chemistry analyses, to Travis May, Curtis Crouse, and Anna DiCocco for fish husbandry and project assistance; to Kata Sharrer for

engineering drawings; to Shanen Cogan and JC Stanley for construction of velocity boosting systems, to Christine Lepine for literature review assistance, to Dr. Brian Vinci for assistance with off-flavor decay modeling, and to Dr. Brett Kenney, Susan Slider, and Sean Estep of West Virginia University's Division of Food Sciences for proximate compositional analyses. This research was funded by the U.S. Department of Commerce/NOAA Sea Grant PTE Federal Award No. NA18OAR4170336 (DOC)/843K592 (UW-Madison), Subaward No. 19–05. Opinions, conclusions, and recommendations are those of the authors and do not necessarily reflect views of the funding body. All experimental protocols complied with Animal Welfare Act (9CFR) requirements and were approved by TCFFI's Institutional Animal Care and Use Committee. The Conservation Fund and the U.S. Department of Commerce/NOAA Sea Grant are equal opportunity employers and providers.

References

Aksnes, A., Gjerde, B., Roald, S.O., 1986. Biological, chemical and organoleptic changes during maturation of farmed Atlantic salmon, Salmo salar. Aquaculture 53, 7–20. https://doi.org/10.1016/0044-8486(86)90295-4.

American Public Health Association (APHA), 2012, Standard Methods for the Examination of Water and Wastewater, 22nd ed. Washington, DC.

AOAC (Association of Analytical Chemists), 1990. Official Methods of Analysis, 15th edition.,. AOAC International, Washington D.C., USA.

Azaria, S., van Rijn, J., 2018. Off-flavor compounds in recirculating aquaculture systems (RAS): production and removal processes. Aquacult. Eng. 83, 57–64. https://doi.org/10.1016/j.aquaeng.2018.09.004.

Burr, G.S., Wolters, W.R., Schrader, K.K., Summerfelt, S.T., 2012. Impact of depuration of earthy-musty off-flavors on fillet quality of Atlantic salmon, Salmo salar, cultured in a recirculating aquaculture system. Aquacult. Eng. 50, 28–36. https://doi.org/10.1016/j.aquaeng.2020.102104.

Davidson, J., Grimm, C., Summerfelt, S., Fischer, G., Good, C., 2020. Depuration system flushing rate affects geosmin removal from market-size Atlantic salmon Salmo salar. Aquac. Eng. 90, 102104 https://doi.org/10.1016/j.aquaeng.2020.102104.

Davidson, J., May, T., Good, C., Waldrop, T., Kenney, B., Terjesen, B., Summerfelt, S., 2016. Production of market-size North American strain Atlantic salmon Salmo salar in a land-based recirculation aquaculture system using freshwater. Aquacult. Eng. 74, 1–16. https://doi.org/10.1016/j.aquaeng.2016.04.007.

Davidson, J., Schrader, K., Ruan, E., Swift, B., Aalhus, J., Juarez, M., Wolters, W., Burr, G., Good, C., Summerfelt, S.T., 2014. Evaluation of depuration procedures to mitigate the off-flavor compounds geosmin and 2-methylisoborneol from Atlantic salmon Salmo salar raised to market-size in recirculating aquaculture systems. Aquacult. Eng. 61, 27–34. https://doi.org/10.1016/j.aquaeng.2014.05.006.

Davison, W., 1997. The effects of exercise training on teleost fish, a review of literature. Comp. Biochem. Physiol. A 117, 67–75. https://doi.org/10.1016/S0300-9629(96)00284-8.

Engle, C.R., Pounds, G.L., Ploeg, M., 1995. The cost of off-flavor. J. World Aquacult. Soc. 26, 297–306. https://doi.org/10.1111/j.1749-7345.1995.tb00258.x.

Gobas, F.A.P.C., MacKay, D., 1987. Dynamics of hydrophobic organic chemical bioconcentration in fish. Environ. Toxicol. Chem. 6, 495–504. https://doi.org/10.1002/etc.5620060702.

HACH Company, 2003. DR/4000 Spectrophotometer Procedures Manual. Hach Company, Loveland, Co.

HACH Company, 2015, HACH Water Analysis Handbook, 8th ed., USA.

Houle, S., Schrader, K.K., Le François, N.R., Comeau, Y., Kharoune, M., Summerfelt, S.T., Savoie, A., Vandenberg, G.W., 2011. Geosmin causes off-flavour in arctic charr in recirculating aquaculture systems. Aquacult. Res. 42, 360–365. https://doi.org/10.1111/j.1365-2109.2010.02630.x.

Howgate, P., 2004. Tainting of farmed fish by geosmin and 2-methyl-iso-borneol: a review of sensory aspects and of uptake/depuration. Aquaculture 234, 155–181. https://doi.org/10.1016/j.aquaculture.2003.09.032.

Intrafish, 2018, Land-based salmon farming: Aquaculture's new reality. Fischer, E. (Ed.).

Jobling, M., Baardvik, B.M., Christiansen, J.S., Jørgensen, E.H., 1993. The effects of prolonged exercise training on growth performance and production in fish. Aquacult. Int. 1, 95–111. https://doi.org/10.1007/BF00692614.

Johnsen, P.B., Lloyd, S.W., 1992. Influence of fat content on uptake and depuration of the off-flavor 2-methylisoborneol by channel catfish (Ictalurus punctatus). Can. J. Fish. Aquat. Sci. 49, 2406–2411. https://doi.org/10.1139/f92-266.

Lindholm-Lehto, P.C., Vielma, J., 2019. Controlling of geosmin and 2–methylisoborneol induced off-flavours in recirculating aquaculture system farmed fish—a review. Aquacult. Res. 50, 9–28. https://doi.org/10.1111/are.13881.

Lloyd, S.W., Grimm, C.C., 1999. Analysis of 2-methylisoborneol and geosmin in catfish by microwave distillation - solid phase microextraction. J. Agric. Food Chem. 47, 164–169. https://doi.org/10.1021/jf980419x.

Lloyd, S.W., Lea, J.M., Zimba, P.V., Grimm, C.C., 1998. Rapid analysis of geosmin and 2-methylisoborneol in water using solid phase micro extraction procedures. Water Res. 32, 2140–2146. https://doi.org/10.1016/S0043-1354(97)00444-2.

May, T., 2020. Purging for profit: Quantifying off-flavor effect on profitability. RASTech Mag. 3 (3), 12–13. ⟨https://mydigitalpublication.com/publication/?m=60230&i=668244&p=12⟩.

Mowi, 2020, Salmon Farming Industry Handbook, 2020. p. 46. ⟨https://corpsite.azureedge.net/corpsite/wp-content/uploads/2020/06/Mowi-Salmon-Farming-Industry-Handbook-2020.pdf⟩.

NASDAQ, 2021, ⟨https://salmonprice.nasdaqomxtrader.com/⟩ (accessed 17 May 2021).

Petersen, M.A., Hyldig, G., Strobel, B.W., Henriksen, N.H., Jørgensen, N.O., 2011. Chemical and sensory quantification of geosmin and 2-methylisoborneol in rainbow trout (Oncorhynchus mykiss) from recirculated aquacultures in relation to concentrations in basin water. J. Agric. Food Chem. 59, 12561–12568. https://doi.org/10.1021/jf2033494.

Randall, D.J., Connell, D.W., Yang, R., Wu, S.S., 1998. Concentrations of persistent lipophilic compounds in fish are determined by exchange across the gills, not through the food chain. Chemosphere 37, 1263–1270. https://doi.org/10.1016/S0045-6535(98)00124-6.

Robertson, R.F., Jauncey, K., Beveridge, M.C.M., Lawton, L.A., 2005. Depuration rates and the sensory threshold concentration of geosmin responsible for earthy-musty taint in rainbow trout, Oncorhynchus mykiss. Aquaculture 245, 89–99. https://doi.org/10.1016/j.aquaculture.2004.11.045.

Schrader, K.K., Acuña-Rubio, S., Piedrahita, R.H., Rimando, A.M., 2005. Geosmin and 2-methylisoborneol cause off-flavors in cultured largemouth bass and white sturgeon reared in recirculating-water systems. N. Am. J. Aquacult. 67, 177–180. https://doi.org/10.1577/A04-070.1.

Schrader, K.K., Summerfelt, S.T., 2010. Distribution of off-flavor compounds and isolation of geosmin-producing bacteria in a series of water recirculating systems for rainbow trout culture. N. Am. J. Aquacult. 72, 1–9. https://doi.org/10.1577/A09-009.1.

Schram, E., Kwadijk, C., Blom, E., Verreth, J.A.J., Murk, A.J., Schram, J.W., 2021. Interactive effects of temperature and water exchange of depuration tanks on geosmin excretion by Atlantic salmon (Salmo salar). Aquaculture 535, 736358. https://doi.org/10.1016/j.aquaculture.2021.736358.

Schram, E., Schrama, J., Kusters, K., Kwadijk, C., Palstra, A., 2016. Effects of exercise and temperature on geosmin excretion by European eel (Anguilla anguilla). Aquaculture 451, 390–395. https://doi.org/10.1016/j.aquaculture.2015.09.032.

Schram, E., van Kooten, T., van de Heul, J.W., Schrama, J.W., Verreth, J.A., Mruk, A.J., 2017. Geosmin depuration from European eel (Anguilla anguilla) is not affected by the water renewal rate of depuration tanks. Aquacult. Res. 48, 4646–4655. https://doi.org/10.1111/are.13287.

Schultz, I.R., Hayton, W.L., 1999. Interspecies scaling of the bioaccumulation of lipophilic xenobiotics in fish: an example of using trifluralin. Environ. Toxicol. Chem. 18, 1440–1449. https://doi.org/10.1002/etc.5620180714.

Solstorm, F., Solstorm, D., Oppedal, F., Ferno, A., Fraser, T.W.K., Olsen, R.-E., 2015. Fast water currents reduce production performance of post-smolt Atlantic salmon Salmo salar. Aquacult. Environ. Interact. 7, 125–134. https://doi.org/10.3354/aei00143.

Streit, B., 1998. Bioaccumulation of contaminants in fish. In: Bruanbeck, T., Hinton, D.E., Streit, B. (Eds.), Fish Ecotoxicology (EXS: 86). Birkhauser, Basel, pp. 353–387.

Summerfelt, S., Christianson, L., 2014. Fish farming in land-based closed containment systems. World Aquac. Mag. 2014, 18–21. March. ⟨https://www.was.org/articles/fish-farming-in-land-based-closed-containment-systems.aspx#.YFzNp9KSlaR⟩.

Timmerhaus, G., Lazado, C.C., Nikko, A.R.C., Reiten, B.K.M., Johansen, L.-H., 2021. The optimum velocity for Atlantic salmon post-smolts in RAS is a compromise between muscle growth and fish welfare. Aquaculture 532, 736076. https://doi.org/10.1016/j.aquaculture.2020.736076.

Totland, G.K., Kryvi, H., Jødestøyl, K.A., Christianson, E.N., Tangerås, A., Slinde, E., 1987. Growth and composition of the swimming muscle of adult Atlantic salmon (Salmo salar L.) during long-term sustained swimming. Aquaculture 66, 299–313. https://doi.org/10.1016/0044-8486(87)90115-3.

Tucker, C.S., 2000. Off-flavor problems in aquaculture. Rev. Fish. Sci. 8, 45–88. https://doi.org/10.1080/10641260091129170.

Waldrop, T., Summerfelt, S., Mazik, P., Good, C., 2018. The effects of swimming exercise and dissolved oxygen on growth performance, find condition, and precocious maturation. Aquacult. Res. 49, 801–802. https://doi.org/10.1111/are.13511.

Yang, R., Brauner, C., Thurston, V., Neuman, J., Randall, D.J., 2000. Relationships between toxicant transfer kinetic processes and fish oxygen consumption. Aquat. Toxicol. 48, 95–108. https://doi.org/10.1016/S0166-445X(99)00050-8.