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The impact of water exchange rate on the health and performance of rainbow trout *Oncorhynchus mykiss* in water recirculation aquaculture systems

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ABSTRACT

A controlled six-month study was conducted in six replicated water recirculation aquaculture systems (WRAS) to assess fish health indicators in relation to high feeding/low flushing conditions. Juvenile rainbow trout (*Oncorhynchus mykiss*) were stocked in six identical WRAS (1000 fish per system), and were maintained over the study period between 25 kg/m^3 (minimum) and 80 kg/m^3 (maximum) densities. Three WRAS received a relatively high makeup water exchange rate (2.6%), while three systems received only 0.26% exchange; mean feed loadings of 0.39 and 4.1 kg/day per m^3/day of makeup water flow were maintained for the high and low exchange treatments, respectively. At the end of the study period, there were surprisingly no significant ($p<0.05$) differences in average fish weight between treatment groups. Percentage survival was excellent in both high and low makeup system populations (99.5% ± 0.1 and 98.9% ± 0.4, respectively). Histopathological evaluation revealed a significant increase in splenic and skin lesions in low makeup system fish relative to those sampled from the high makeup systems, while lesions in the posterior kidney were more prevalent in the high makeup system fish. Plasma chloride and blood urea nitrogen were also significantly different between fish from the two treatment groups, and caudal fin quality assessment showed significantly greater fin erosion in low makeup system fish. Despite these clinical and subclinical differences, there appeared to be no major treatment effect on overall fish performance.

1. Introduction

As the relative amount of new water (i.e. makeup water) entering a water recirculation aquaculture system (WRAS) decreases, system flushing is reduced and water quality within the system is consequently degraded. Observations at The Freshwater Institute in 2004 indicated that during conditions of high feeding and low flushing in WRAS (i.e. 1.3–2.0 kg feed/day per m^3/day of makeup water flow), a corresponding decline in rainbow trout *Oncorhynchus mykiss* health (i.e. increased mortality) of unknown etiology occurs. During this mortality episode, all tested water quality parameters were within safe limits, the WRAS was assessed to be operating satisfactorily, and no infectious disease problems were diagnosed or suspected as causes of the mortality.

This mortality trend could represent an important barrier to operating WRAS with high feeding rates during periods of limited makeup water availability, and therefore recent research at The Freshwater Institute has been carried out to achieve a better understanding of environmental and fish health changes in WRAS during conditions of high feeding and low flushing. Because the cause of the previously observed fish health decline was unknown, data on a wide range of fish health indicators were collected in the present study to enhance the possibility of identifying important variables associated with mortality in high feeding and low flushing WRAS.

2. Materials and methods

Beginning in January 2007, a controlled six-month study was conducted in six replicated WRAS to assess fish health indicators in relation to high feed and low flushing conditions. Each WRAS consisted of the following components: fluidized-sand biofilter, CO_2 stripping column, low head oxygenation (LHO) unit, circular dual drain culture tank (5.3 m^3), radial flow settler, microscreen drum filter (60 μm), heat exchanger, and a 1-HP centrifugal pump. Total system volume was 9.5 m^3 with a recirculation rate of 380 L/min (100 gpm). Fig. 1 illustrates the basic arrangement of an individual system's components. In addition to the six experimental WRAS, three small (0.5 m^3) circular tanks within a flow-through system in a separate building were used as comparison tanks for fish performance during the study (no 5.3 m^3 flow-through tanks were available for comparison, but fish were maintained in these smaller tanks at identical densities and feeding rates as the six experimental WRAS).

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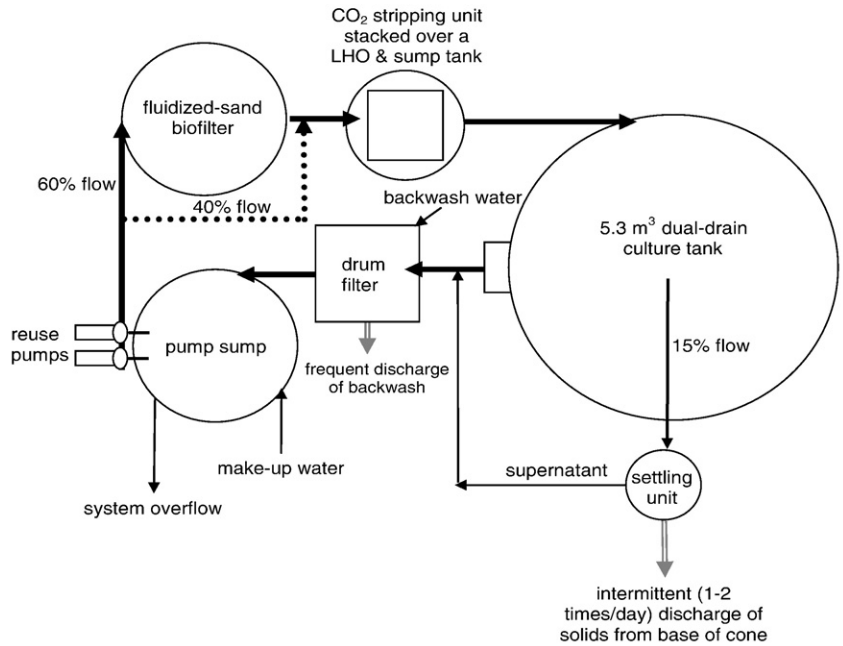

Fig. 1. Summary diagram of an individual experimental water recirculating aquaculture system employed in this study.

Rainbow trout *O. mykiss* (acquired as eyed eggs from a commercial producer from a single spawn, and incubated and raised on site at The Freshwater Institute) were maintained for 6 months at a density range of 25 kg/m^3 to 80 kg/m^3. Approximately 1000 rainbow trout were stocked (averaging 133 ± 1 g) into each WRAS, and 100 fish from the same cohort were stocked into each of the three flow-through comparison tank. Three WRAS received a high makeup water exchange rate (2.6%), while three systems received a low 0.26% exchange. Exchange rates were set on a flow basis (i.e., for high exchange (2.6%) WRAS, the 100-gpm system recirculation flow contained 2.6 gpm of makeup water), and high and low exchange treatments were randomly allocated among the six WRAS. All WRAS makeup water, as well as water supplying the flow-through comparison tanks, originated from a spring with approximately 1200 gpm flow. Mean feed loadings of 0.39 and 4.1 kg/day per m^3/day of makeup water flow were maintained for the high and low treatments, respectively. Feeding was estimated based on standardized feeding charts, as well as observations of feeding activity and wasted feed. A constant 24-hour photoperiod was provided, and all tanks were fed equal portions once every 2 h using automated feeders (T-drum 2000CE, Arvo-Tec, Finland). By controlling the rearing environment in this way, constant biological respiration and waste production rates were produced.

Water quality sampling was conducted weekly throughout the study, and measured parameters included total ammonia nitrogen, unionized ammonia, nitrite, nitrate, alkalinity, dissolved oxygen, carbon dioxide, biochemical oxygen demand, true color, ultraviolet transmittance, total suspended solids, total particles, phosphorus, temperature, total organic carbon, dissolved organic carbon, and pH. The water quality component of this investigation is only briefly summarized in this paper; for a complete description of the methodologies and findings of the water quality analyses please refer to Davidson et al. (2008) and Davidson et al. (in press).

To assess performance, length and weight data were collected on a monthly basis, from which growth curves were generated, feed conversion was assessed, and thermal growth coefficients were calculated using the following formula (Iwama and Tautz, 1981):

$$\left[\mathrm{FBW}^{1/3} - \mathrm{IBW}^{1/3}\right] / \sum [T \times D] \times 100$$

where FBW = final body weight; IBW = initial body weight; T = water temperature; and D = number of days.

Calculated sample sizes for performance data collections ranged from 50 to 120 fish, depending on estimated weight variation in the population (a 90% confidence was used in the sample size calculations, with an acceptable error of 5 g). Mortalities were also noted on a daily basis, and percentage survival was calculated for each treatment at the study's end using the following formula: [(initial fish number − mortalities)/initial fish number]*100.

Samples of nine separate tissues were collected from five fish per tank at the beginning of the study, and approximately every 2 months thereafter until the study's end, and were preserved in histological grade 10% formalin solution (Fisher Scientific) for at least 1 week prior to shipment to the Washington Animal Disease Diagnostic Laboratory (Pullman, WA) for processing and histopathological evaluation. The sampled tissues included portions of skin, gill, heart, liver, spleen, pyloric cecae, swim bladder, and anterior and posterior kidney, and all tissues were sampled at the same anatomical region (e.g. all gills were sampled on the left side, taking the middle third of the second gill arch). The pathologist conducting the evaluations was blinded as to which treatment group each sampled fish had been allocated. Qualitative observations regarding tissue pathology were carried out for all sampled organs, with additional grading of gill lesions according to the Snekvik Grading Scale (Grades 0 through 4) (Dr. Kevin Snekvik, Washington Disease Diagnostic Laboratory, pers. comm.). Grade 1 inflammation denoted mild epithelial hyperplasia involving less than 10% of the evaluated section, in addition to mild levels of inflammatory infiltrates in the primary and secondary lamellae, while Grade 3 inflammation reflected moderate epithelial hyperplasia with fusion of secondary lamellae involving 25–50% of the examined gill section, as well as moderate inflammatory infiltrates in the primary and secondary lamellae (gill lesions observed in this study were not graded higher than Grade 3).

In general, due to the difficulty in creating a reliable, quantifiable scale for the qualitative pathological observations, it was expedient to initially combine all lesion types observed for each tissue and create a dichotomous outcome variable denoting a general presence or absence of lesions noted within each tissue evaluated. With this outcome, bivariable logistic regression models were created using STATA 9 software (StataCorp, College Station, TX) to assess the

Table 1
Summary of weekly water quality assessments, measured at WRAS tank side drain outlets, over the six-month study period.

Water quality parameters	High exchange WRAS	Low exchange WRAS
Total ammonia nitrogen (mg/L)*	0.29 ± 0.00	0.48 ± 0.05
Unionized ammonia (mg/L)*	0.002 ± 0.000	0.004 ± 0.000
Nitrite nitrogen (mg/L)*	0.04 ± 0.01	0.10 ± 0.00
Nitrate nitrogen (mg/L)*	12 ± 0	70 ± 4
Alkalinity (mg/L)*	226 ± 1	214 ± 1
Dissolved carbon dioxide mg/L)	9 ± 0	9 ± 0
Carbonaceous biochemical oxygen demand (mg/L)*	2 ± 0	6 ± 1
True color (Pt–Co units)*	11 ± 1	74 ± 9
Ultraviolet transmittance (%)*	89 ± 0	53 ± 4
Phosphorous (mg/L)*	0.52 ± 0.01	3.11 ± 0.22
Total suspended solids (mg/L)*	2.7 ± 0.1	6.4 ± 1.2
Total particles (0–60 µm)*	6618	13,849
Temperature (°C)	13.2 ± 0.0	13.2 ± 0.1
Total organic carbon (mg/L)*	3.93 ± 0.29	13.04 ± 0.88
Dissolved organic carbon (mg/L)*	3.28 ± 0.25	10.42 ± 0.67
pH	7.53 ± 0.03	7.54 ± 0.04
Dissolved oxygen (mg/L)	10.0 ± 0.0	10.0 ± 0.1

* Indicates significant ($p<0.05$) difference between treatment groups.

Table 2
Bivariable logistic regression models comparing the general presence or absence of lesions within specific tissues noted in fish reared in low exchange versus high exchange recirculating aquaculture systems (all sampling data combined).

Tissue	Odds ratio	95% confidence interval	*p*-value
Gill	0.48	(0.16, 1.41)	0.179
Liver	1.64	(0.61, 4.44)	0.327
Heart	1.00	(0.45, 2.21)	1.000
Spleen	1.57	(1.02, 2.42)	0.042
Anterior kidney	0.48	(0.17, 1.34)	0.162
Posterior kidney	0.34	(0.21, 0.57)	<0.000
Swim bladder	–	–	–
Skin	2.92	(1.76, 4.85)	<0.000

relationship between treatment groups and observed tissue pathology. These regression analyses were repeated to compare fish in WRAS (both treatment groups combined) with fish reared in the flow-through comparison tanks, using combined data from all sampling events conducted throughout the study period. To further elucidate the nature of the pathologies detected in all fish examined, prevalences of each pathology type within each tissue were calculated for fish in high exchange WRAS, low exchange WRAS, and flow-through comparison tanks. These prevalences were calculated using data from the final two sampling events only, as analyses of results obtained from individual sampling events revealed that significant differences in tissue pathology between treatments only occurred towards the end of the study period (i.e. during the period of the final two samplings), and were not present throughout approximately the first two-thirds of the study (results not shown).

At the end of the six-month study period, five fish per tank were euthanized with an overdose of Tricaine-S (tricaine methanesulfonate; Western Chemical, Inc.) and 3 ml blood samples were immediately collected via caudal venipuncture. These blood samples were quickly centrifuged, and the separated plasma was collected, frozen, and shipped to Cornell University's Animal Health Diagnostic Center (Ithaca, NY) for small animal plasma chemistry profile analysis. Reported data were compiled and imported into STATA, and as they were assessed to be non-parametric a Kruskal–Wallis rank test was employed to determine significant differences between the treatment groups.

Finally, at the study's end it was noted that fin quality had generally deteriorated. To determine whether there were differences in fin erosion between treatment populations, approximately 110 fish per tank (WRAS only) were sampled, and caudal fin quality was subjectively evaluated through non-blinded assessments. Tails were graded as being in good, fair, or poor condition. Prevalences for each tail condition within each treatment group were calculated, and were compared statistically using ANOVA testing.

3. Results

3.1. Water quality

Within 24 h of study initiation, water was observed to be qualitatively darker and more turbid in low makeup water exchange WRAS relative to the high makeup exchange systems, and these obvious visible differences were persistent throughout the study period. The majority of water quality parameter means were significantly ($p<0.05$) different between the high and low makeup water exchange groups (Table 1). All water quality parameters measured in either treatment group, however, were within published acceptable ranges for safe salmonid rearing conditions (Piper et al.,

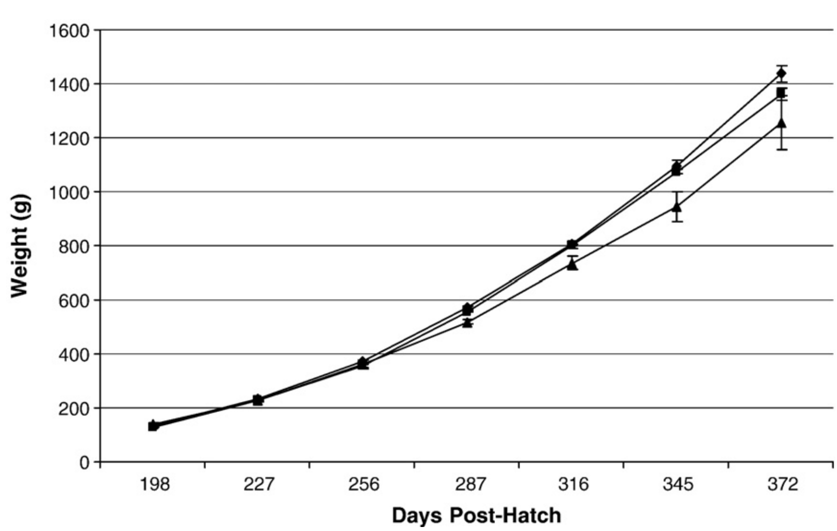

Fig. 2. Fish growth curves for the six-month study period, for high exchange (◆), low exchange (■), and flow-through comparison (▲) groups.

Table 3
Bivariable logistic regression models comparing the general presence or absence of observed lesions within specific tissues in fish reared in recirculating aquaculture systems (both high and low exchange treatments combined) versus flow-through comparison tanks.

Tissue	Odds ratio	95% confidence interval	*p*-value
Gill	1.64	(0.70, 3.84)	0.254
Liver	0.52	(0.23, 1.16)	0.111
Heart	2.11	(1.44, 3.10)	<0.000
Spleen	2.47	(1.56, 3.90)	<0.000
Anterior kidney	0.74	(0.20, 2.73)	0.647
Posterior kidney	0.76	(0.22, 2.60)	0.667
Swim bladder	2.03	(0.24, 17.56)	0.513
Skin	1.92	(0.60, 6.15)	0.274

1982; Brune and Tomasso, 1991; Losordo, 1991; Heinen, 1996; Danley et al., 2005; Colt, 2006).

3.2. Fish performance

No statistically significant differences in mean fish weight between treatment groups were detected during the entire six-month study period (Fig. 2). The overall mean final weight was 1377 g after 6 months when the fish were 372 days post-hatch in age; for high exchange fish, the final mean weight was 1401 ± 23 g, while low exchange fish finished the study with a mean weight of 1366 ± 33 g. Fish held in the flow-through comparison tanks had a mean final weight of 1254 ± 100 g, which was not significantly different from either treatment group reared in the WRAS. Feed conversion ratios (FCR) were not significantly different between the high and low exchange groups (1.38 ± 0.11 and 1.45 ± 0.11, respectively), although the FCR for the flow-through comparison tanks (1.95 ± 0.17) was significantly higher than either WRAS treatment group. Thermal growth coefficients were comparable among all study groups, with mean values for the high exchange, low exchange, and flow-through comparison tanks calculated as 2.64 ± 0.13, 2.62 ± 0.17, and 2.56 ± 0.07, respectively. These thermal growth coefficient values were similar to other cohorts of rainbow trout raised at The Freshwater Institute. Final survival values were high in all treatment groups: $99.5\% \pm 0.1$, $98.9\% \pm 0.4$, and $99.3\% \pm 0.0$ for the high exchange, low exchange, and flow-through comparison groups, respectively. A small but statistically significant difference ($p = 0.0495$) in survival was detected between the high and low exchange groups; however, sample size was small ($n = 3$) and due to the non-normality of the survival data, a non-parametric (Kruskal–Wallis) test was employed. With this in mind, we can only conclude that the low exchange environment caused slightly increased mortality, but survival was still high in this population.

Table 5
Mean plasma chemistry values of selected parameters for fish in high exchange WRAS, low exchange WRAS, and flow-through comparison tanks, with samples taken at the end of the six-month study period.

Parameter	Treatment	Mean	Standard error	Statistical significance
Sodium (mEq/L)	High exchange	157.1	0.84	
	Low exchange	157.9	0.83	
	Flow-through	159.3	1.06	
Potassium (mEq/L)	High exchange	2.37	0.84	B
	Low exchange	2.41	0.83	
	Flow-through	1.73	1.06	
Chloride (mEq/L)	High exchange	124.1	0.80	A, B
	Low exchange	131.0	1.24	
	Flow-through	122.1	1.04	
Urea Nitrogen (mg/dL)	High exchange	15.9	0.62	A, B
	Low exchange	19.0	0.80	
	Flow-through	<2.0	0.00	
Glucose (mg/dL)	High exchange	78.1	5.27	B
	Low exchange	76.4	6.99	
	Flow-through	88.1	4.60	

A = significant ($p<0.05$) difference between high and low exchange WRAS; B = significant ($p<0.05$) difference between recirculating systems (high and low exchange WRAS combined) and flow-through comparison tanks.

3.3. Histopathology

Results of the bivariable logistic regression modeling indicated that both spleen and skin tissues were significantly more likely to exhibit histopathological lesions in fish reared in low exchange systems than those raised in high exchange systems. Conversely, posterior kidney pathology was less likely to be observed in fish from low exchange WRAS relative to the high exchange systems (Table 2). Odds ratios reported represent the relative odds of having lesions observed in a specific tissue from a low exchange versus a high exchange WRAS (Odds ratios greater than one reflect higher odds of having lesions noted from organ samples from a low exchange system, while odds ratios less than one reflect lower relative odds of pathology observed in organ samples collected from low exchange system fish). Fish in recirculating systems (regardless of makeup exchange rate) were significantly more likely to exhibit pathology in heart and splenic tissues relative to their counterparts in flow-through tanks (Table 3).

Actual lesions observed in each tissue type collected during the final two sampling events are summarized in Table 4. The prevalence of each specific lesion was generally low, and therefore statistical testing was not carried out due to incomplete distribution of

Table 4
Specific histopathological lesions noted in each tissue examined in the final two sampling events of the six-month study, with prevalences of each tissue lesion for high exchange, low exchange, and flow-through comparison treatments.

Tissue	Lesion	Prevalence in treatment group		
		High exchange	Low exchange	Flow-through
Gill	Lymphocytic branchitis, Grade 1	27% (8/30)	40% (12/30)	40% (12/30)
	Lymphocytic branchitis, Grade 2	7% (2/30)	7% (2/30)	–
	Lymphocytic branchitis, Grade 3	3% (1/30)	–	3% (1/30)
Liver	Lipidosis	3% (1/30)	23% (7/30)	20% (6/30)
	Lymphocytic hepatitis	17% (5/30)	30% (9/30)	33% (10/30)
Heart	Lymphocytic epicarditis	3% (1/30)	17% (5/30)	3% (1/30)
Spleen	General congestion	23% (7/30)	37% (11/30)	7% (2/30)
Anterior kidney	Nephrocalcinosis	3% (1/30)	–	–
Posterior kidney	Nephrocalcinosis	7% (2/30)	–	–
	Granuloma	7% (2/30)	–	–
Swim bladder	Lymphocytic aerocystitis	13% (4/30)	–	–
Skin	Lymphocytic epidermitis	7% (2/30)	17% (5/30)	7% (2/30)
	Lymphocytic dermatitis	–	7% (2/30)	–
	Lymphocytic myositis	–	3% (1/30)	3% (1/30)

Table 6
Prevalences of caudal fins subjectively assessed to be in good, fair, or poor condition within high and low exchange treatment groups.

Caudal fin condition	Prevalence (%)		
	High exchange	Low exchange	*p*-value
Good	86.8	63.1	0.0012
Fair	7.8	18.9	0.0070
Poor	5.4	18.0	0.0108

individual lesion types among all replicated units within each treatment population. In general, higher prevalences of each specific lesion were observed in samples from low exchange treatment fish, although kidney lesions and swim bladder inflammation were only observed in fish from the high exchange group. Inflammation noted in the various tissue types was largely lymphocytic in nature.

3.4. Plasma chemistry

With the exception of blood urea nitrogen, all plasma chemistry parameters measured (Table 5) were within published normal ranges for rainbow trout (Stoskopf, 1993; Wedemeyer, 1996). No differences in plasma sodium were detected between treatments groups. Both plasma chloride and blood urea nitrogen were significantly higher in low exchange treatment fish relative to high exchange fish. Plasma potassium, chloride, and blood urea nitrogen were significantly higher in fish reared in WRAS (both treatment groups combined) relative to those raised in the flow-through comparison tanks. Flow-through fish demonstrated higher plasma glucose levels relative to both WRAS treatment groups.

3.5. Fin condition

After 6 months' exposure to the different makeup water exchange rates, poor fin condition was significantly associated with low exchange systems, while fish assessed to have good quality tails were significantly more prevalent in the high makeup water exchange WRAS (Table 6). Representative illustrations of fin condition are shown in Fig. 3.

4. Discussion

The previously observed decline in fish health in low exchange WRAS at The Freshwater Institute was not reproduced in this study, despite the significant differences in water quality between the high and low makeup water exchange treatment groups (Davidson et al., 2008; Davidson et al., in press). There were surprisingly no statistically significant differences in fish size at the end of the six-month study period. Fish held in the flow-through comparison tanks were smaller (though not significantly so) than fish in either WRAS treatment group, and this size difference was most likely related to differences in tank dimensions relative to the WRAS tanks (i.e. due to their much smaller water depth, feed in the flow-through tanks was able to fall to the bottom drain in less time than in the larger WRAS tanks; FCR data supports the possibility that more feed was lost in these tanks relative to the WRAS). Survival in each treatment group was high. While occasional opportunistic infections (*Flavobacterium branchiophilum*-associated bacterial gill disease, and mild *Ichthyobodo necator* (costia) infestations) were observed through wet-mount gill and skin preparations from mortalities and culls throughout the course of the study, these tended to be observed on individual fish only, and no actual population-level outbreaks associated with opportunistic infections were experienced. Furthermore, these opportunistic pathogens were observed in fish from all WRAS tanks regardless of makeup exchange rate (although no pathogens were noted on fish from the flow-through comparison tanks), and no clear pattern was determined in the presence of opportunistic pathogens on mortalities and culls between high and low exchange treatment groups.

It is interesting to note that low-level gill inflammation (Grade 1) was higher in prevalence in the low exchange versus the high exchange groups (40% vs. 27%, respectively), but that an equally high prevalence was also found in fish from the flow-through comparison tanks. While general gill inflammation is often associated with reduced water quality (Ferguson, 1989), particularly with elevated levels of total suspended solids (as observed in the low exchange WRAS tanks), fish in the flow-through comparison tanks were exposed to relatively clean water, and hence, in the absence of any infectious etiology it is unknown why the prevalence of low-level inflammation was similar in these tanks to the low exchange WRAS. Furthermore, while very low in prevalence (3%), the highest grade of gill inflammation (Grade 3) was only noted in the relatively clean waters of the high exchange and flow-through tanks. The exact causes of the observed gill pathology are therefore unlikely to be related to reduced water quality, and require further investigation.

Because lymph drains into the pericardial sac surrounding the heart, the heart's external surface (epicardium) can often exhibit pathology related to parasitic and bacterial agents (Ferguson, 1989) causing systemic infection. Total heterotrophic bacteria counts in the low makeup exchange WRAS (results not shown) were approximately five times greater than in the high makeup exchange systems, and approximately 30 times greater than in the flow-through tanks (Davidson et al., 2008), and therefore the observed higher prevalence of epicarditis in the low makeup systems could, in general, be related to the greater bacterial challenge with persistent, relatively increased antigen exposure experienced by fish in these systems. Furthermore, as the spleen is one of the major organs responsible for filtering the vascular system, the increased splenic congestion observed in fish in low exchange systems (and WRAS in general relative to the flow-through comparisons) might also indicate a higher degree of immune system challenge related to higher bacterial counts in the water, and possibly to consequent bacteremia.

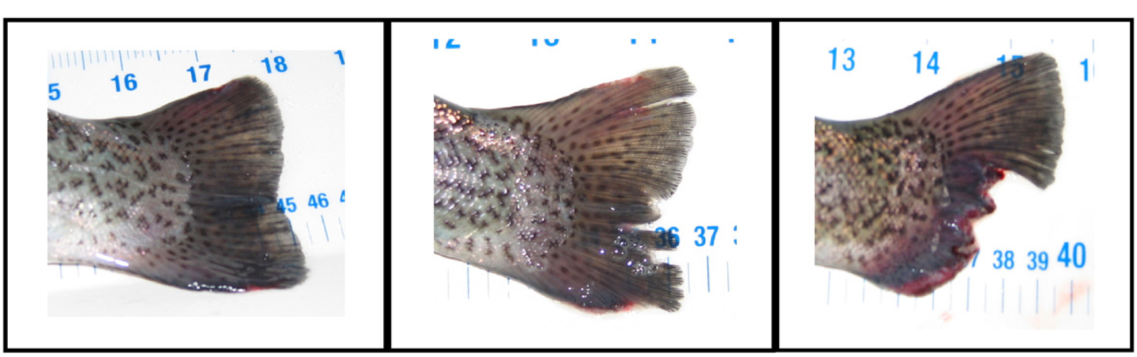

Fig. 3. Caudal fin erosion: examples of tails subjectively categorized as being in (left to right) good, fair, and poor condition.

The epidermis was also shown to be an area of increased inflammation in fish in low makeup WRAS relative to those in high makeup WRAS or flow-through tanks. The recruitment of lymphocytes to the skin's outer layer reflects an environmental, on-going immunogenic stimulation. Whether this recruitment is caused by physical (e.g. total suspended solids or particles) irritation or by bacterial stimulation in low exchange WRAS is unknown, and further research in this area would be beneficial given the skin's importance in fish as a barrier to opportunistic infections and to the external environment in general (Stoskopf, 1993).

Plasma potassium levels were generally lower than those previously published (3.2–3.5 mEq/L; Witters, 1986) for healthy rainbow trout; however, normal ranges for plasma chemistry parameters are variable depending on many rearing environment factors, and there is still a need for more published data in this area (Wedemeyer, 1996). Observed chloride levels were within published normal limits (Meade and Perrone, 1980; Wedemeyer, 1996), but as with potassium, lower levels were found in fish exposed to relatively clean water (flow-through tanks versus WRAS for potassium and chloride, and high versus low exchange WRAS for chloride). Both electrolytes tend to be lower in the blood of fish with generalized infections (Stoskopf, 1993), and hypochloremia can be detected in fish exposed to stressful stimuli (Wedemeyer et al., 1990) due to a net loss of chloride ions through the gills. While it is unlikely that the relatively low plasma electrolyte levels observed in this study were due to systemic infections (no morbidity was noted in sampled fish), it is possible that fish reared in the flow-through tanks, either through water quality/clarity- or facility-related factors, were more susceptible to sampling stress, particularly in light of the relatively high plasma glucose detected in these fish. Further investigation measuring plasma cortisol and lactate is needed to supplement this hypothesis, and at present it is unknown why lower electrolyte levels were detected in flow-through tank fish.

In both high and low exchange WRAS, plasma urea nitrogen was elevated above published upper limits (Warner et al., 1978; Stoskopf, 1993; Wedemeyer, 1996) of healthy rainbow trout. Elevated plasma urea nitrogen levels are thought to be associated with liver or gill dysfunction (Stoskopf, 1993), as these are the sites of urea production and excretion, respectively. However, the prevalence of gill and liver inflammation in flow-through comparison fish was similar in the final two histology samplings to fish in the low exchange WRAS, and therefore it is unlikely that pathology in these two organs was responsible for the elevated plasma urea levels. The exact cause of these abnormally high plasma urea levels remains unknown.

Poorer fin condition was associated with low exchange systems. Fin erosion in farmed fish is a complex, multifactorial process (Ellis et al., 2008), with many factors including opportunistic infections (Turnbull et al., 1996), stress (Kindschi et al., 1991), and water quality (Bosakowski and Wagner, 1994) considered to be primary or secondary causes. While it is unknown at present which factor(s) were responsible for the observed differences in fin quality between the two treatments, this finding, taken in context with associated welfare and product quality considerations, should be kept in mind by producers rearing fish in similar low exchange WRAS.

5. Conclusions

We were unable to reproduce the fish health decline previously observed at The Freshwater Institute in WRAS during conditions of high feeding and low flushing. Surprisingly, fish grew very well in all groups in both experiments, despite various statistically significant clinical (fin condition) and subclinical (histopathology, plasma chemistry) differences in fish health parameters between treatment groups. Further research is required to elucidate these findings. In the meantime, current experimentation is being performed and future studies are being planned to continue investigation of fish health issues in WRAS managed at low makeup water exchange rates.

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