

# BOTTLENECKS TO SURVIVAL EVALUATING THE EFFECTIVENESS OF COWICHAN RIVER STEELHEAD (ONCORHYNCHUS MYKISS) KELT RECONDITIONING

Prepared by Jamieson Atkinson, Jeramy Damborg, Sam James

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# **EXECUTIVE SUMMARY**

The study aimed to assess the effectiveness of a reconditioning treatment in improving the condition factor of Cowichan River steelhead kelts as part of an intensive tagging study. The study also assessed the viability of kelt reconditioning as a method for increasing the marine survival of kelts (i.e. increasing the rates of iteroparity). This study was conducted at the Freshwater Fisheries Society's Trout Hatchery in Duncan, B.C. A total of 20 steelhead kelts were captured via angling techniques in 2021, with 13 surviving the 30-day reconditioning treatment; contrary to expectations, the reconditioning did not improve the overall condition of steelhead kelts with the overall condition found to be significantly lower at release than upon arrival. Further, no reconditioned kelts returned to spawn in subsequent years. The process involved high financial costs and logistical challenges due to extensive handling and transportation. Compared to other successful programs, whose goal is to increase steelhead survival and rates of iteroparity, in the Columbia and Yakima rivers, the lack of necessary infrastructure in the Cowichan River limited this study's effectiveness. Given these findings, the report underlines the need for a downstream capture facility, longer holding times, and a reassessment of the program's scale and approach to enhance future outcomes. The current method proved ineffective for increasing steelhead kelt condition before an intensive tagging procedure and indicates a larger program to increase kelt iteroparity is likely to be unsuccessful.

Citation:

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### INTRODUCTION

Anadromous rainbow trout, (*Oncorhynchus mykiss*), or steelhead, are native to coastal and interior streams of Alaska, British Columbia (BC), Washington, Idaho, Oregon, and California (Kendall et.al. 2017). Steelhead are a sought-after game fish. On the Cowichan River, steelhead typically provide around 5,000 angler days per year and can provide up to nearly 12,000 angler days per year (Province of B.C. 2021). The Cowichan River, near Duncan BC, is home to a 'winter-run' ecotype, which typically returns between December and March after 2-3 years at sea. Steelhead spawn between February and April and can reach sizes of more than 9 kg (McPhail et al. 1994). Unlike most Pacific salmon, steelhead are iteroparous, meaning they can spawn multiple times (often 1-2 years following their first spawning season) before perishing (Robards and Quinn 2002). Iteroparity is estimated to be ~10% of the population, but can be highly variable between years and systems (i.e. range from 0.4% to 21%; Clemens 2015). The post-spawn fish, known as kelts, head back to the ocean in the late spring to grow, recondition, and return to spawn a second or, on rare occasions, even a third time (Hatch et al. 2013).

Steelhead abundance has declined significantly over their entire range, with local extirpations in some streams (Ward 2000; Kendall et al. 2017). Many winter-run populations on Vancouver Island, BC, remain well below 25% of historical abundance estimates (M. McCulloch, WLRS Anadromous Fisheries Specialist. Pers, Comm. 2024). The Cowichan River remains one of the few steelhead populations on Vancouver Island that is healthy and can support an in-river sport fishery; populations are estimated to be in the "Routine Management Zone" with annual escapements estimated at 500-1000 fish in recent years (M. McCulloch, WLRS Anadromous Fisheries Specialist. Pers, Comm).

Kelt reconditioning is a novel technique hypothesized to increase overall spawning abundance by increasing the health and condition of the kelts between spawn events (BPA 2016; T. Davies, WLRS Provincial Stock Assessment Scientist. Pers. Comm. 2019). Reconditioning of steelhead kelts requires the capture of kelts as they out-migrate, and treating, holding, and feeding them in an artificial rearing environment to improve the regeneration of gonads and overall survival to repeat spawning (Hatch et al. 2013). To recondition kelts, they can be captured after naturally spawning in the river, transported to a holding facility (i.e. hatchery), medicated, fed and held for several months. This process aims to raise the condition factor of the steelhead kelts to a state where they are healthy and more likely to survive to spawn again (BPA 2016).

The primary objective of this study was to utilize reconditioning methods on steelhead kelts from the Cowichan River to try to increase their condition for a steelhead kelt marine migration study (Davies et al. in prep). This larger study utilized MiniPat satellite tags on angled steelhead kelts, captured in their rivers and released in the ocean after tagging. The study's objectives were to understand the mechanisms of mortality, dispersal, and survival (Photo 4, Appendix A). For full details on this study and its results, please refer to Davies et al. 2024. A secondary objective of the reconditioning trails was to determine the feasibility of conducting a large-scale reconditioning program.

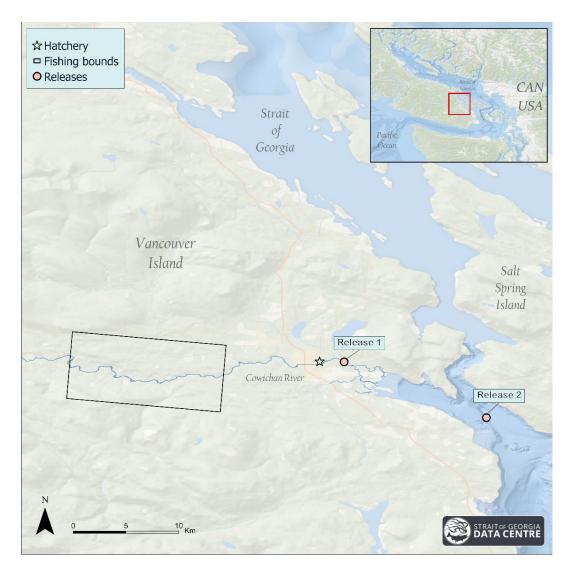
Upon the provincial government's suggestion, the Bottlenecks to Survival Project, a partnership between the Pacific Salmon Foundation (PSF) and the British Columbia Conservation Foundation, explored the viability of using this technique to increase wild spawner abundance in the Cowichan River, BC.



### **METHODS**

#### Kelt Collection and Sampling

Beginning in late March, using standard sport drift fishing techniques, kelts were angled from the Cowichan River between Stoltz Pool (river kilometer (RKM) 27) and Vimy Road (RKM 13) from a drift boat (Figure 1). Captured fish were netted (with a knotless catch and release type landing net) and visually inspected to determine if they were a kelt. Kelts were identified as steelhead with low condition factor (skinny), flaccid/empty belly for females, and often wear on their lower caudal fin. Females were preferentially selected as they are more likely to return to spawn (Hatch et al. 2013).



**Figure 1.** Map of the Cowichan River study area showing river release and ocean release points.

Any fish determined to be in acceptable condition (free of fungus and open wounds or other injuries, including significant hooking injuries) was placed in temporary transport tank (~100 L) on the drift boat with supplementary aeration (1.5v Marine Metal Aerator "box bubbles") and transported to the nearest road access site. Kelts were moved into a small tank truck filled with fresh river water (600 L) for transport to the Freshwater Fisheries Society of BC's Vancouver Island Trout Hatchery site (VITH) on Boys Road, Duncan B.C (Photo 1, Appendix A), near RKM 5.

#### Reconditioning

Upon arrival at the hatchery, the kelts were transitioned from the live-well into an anesthetic bath using Eugenol (clove oil) for bio-sampling (Photo 2; Appendix A). Kelts were measured for fork length (mm), weight (kg), maximum circumference (girth) in front of the dorsal fin (mm), and sex (m/f; visually determined). Individuals were then photographed and tagged with a sterile 12 mm HDX B PIT tag inserted into their body cavity, and any comments regarding the condition were recorded.

Upon arrival, kelts were injected once with Liquamycin (LA) 200. The antibiotic containing 200 mg of oxytetracycline per mL was injected into each fish's dorsal sinus. Each kelt received 0.1 ml/kg of oxytetracyclene to facilitate recovery from disease or fungus before being transferred to their reconditioning tanks.

After bio-sampling, the kelts were placed in a 5 m wide, 1.5 m deep circular tank with a blackout lid for reconditioning (Photo 3, Appendix A). Kelts in the tank were checked daily. Kelts received a 1% body weight per day ration of Skretting BioBrood 6.0mm pellets. Additionally, hand-feeding of kelts with Hikari Bio-Pure Krill was conducted each morning to stimulate a feed response. For this feeding method, the amount of krill fed was not weighed; kelts were fed until they stopped showing interest in the krill. Additional unweighted krill was also mixed into the artificial Skretting diet, which was placed in belt feeders to provide food throughout the day.

The tank was cleaned regularly to remove extra food/fecal matter for the duration of the holding period. Kelts were held and fed in this manner until released on April 20, 2021 (approximately 1 month, Photo 5 and 6, Appendix A). Prior to release, bio-sampling was repeated (i.e. fork length, weight, and girth measured) to determine a change in overall condition.

#### Data Analysis

All data wrangling, graphing and analyses were conducted in R studio (R Core Team. 2021). Data wrangling and cleaning were conducted using the dplyr package (Wickham et al. 2021), analysis was conducted using Base R functions, and all graphs and figures were developed using ggplot2 (Wickham et al. 2019). The linear mixed effects model was developed using the lme4 package (Bates et al. 2015).

#### Condition Factor

Fultons Condition Factor (K) was used to compare overall condition and calculated according to the Htun-Han (1978) equation:

 $K = (W*10^5 \times 100)/L^3$ 

where, W=weight of fish (kg), and L=length of fish (cm).

The biological data gathered at the hatchery's initial 'arrival' and 'release' back into the wild and overall condition were compared to determine if holding kelts had a biologically significant benefit.

#### Linear Regression Analysis

To understand the influence of reconditioning on each fish, we employed a linear mixed effects model (LMM) to accommodate the variability within our dataset. An LMM facilitates the partitioning of variance into fixed effects, attributed to the reconditioning treatment (before and after reconditioning), and random effects, accounting for individual differences among the fish. The model included the condition of the kelts as a fixed effect and fish as a random effect to capture the variation in responses to the reconditioning process across individuals. The model also assessed the duration (days) of the reconditioning treatment.

K Factor<sub>ii</sub> = 
$$60+61$$
×Condition<sub>ij</sub>+  $62$ ×DaysReconditioned<sub>ij</sub> +  $u_i+\epsilon_{ij}$ 

where: 60 is the intercept, representing the average condition value of the fish upon arrival when the effects of other variables are not considered. 61 is the coefficient for the condition effect, quantifying the change in the response variable when moving from the "Arrival" condition to the "Tag" (release) condition. *Condition<sub>ij</sub>* is a categorical fixed effect with two levels: "Arrival" and "Tag", indicating the stage of measurement. 62 is the coefficient for the days reconditioned, where *DaysReconditioned<sub>i</sub>* is a continuous fixed effect representing the number of days the  $i^{th}$  fish was reconditioned before the measurement. And *ui* represents the random effects. This term allows for correlation within fish measurements across the two conditions, assuming that these random effects are normally distributed with a mean of zero and variance  $\sigma u2$ .  $\epsilon_{ij}$  is the residual error term for the  $i^{th}$  fish at the  $j^{th}$  condition, assumed to be normally distributed with mean zero and constant variance  $\sigma^2$ , accounting for random error not explained by the model.

# RESULTS

In total, 20 steelhead (16 females and 4 males) were captured between March 18–31, 2021, and transported to the hatchery for reconditioning. Seven perished in holding, leaving 13 kelts (12 females and 1 male) for release (Table 1; Appendix B). On average, kelt length increased by 7 mm (range 0–18 mm) between arrival and release (Table 1; Figure 2). Weight decreased by 0.21 kg (range –0.84–0.24 kg) between arrival and release, with only 3 of the 13 kelts gaining weight. Circumference or girth decreased on average by 1.2 cm (range –35–55 cm).

#### Reconditioning

A linear mixed effect model (LMM) revealed that condition significantly declined during reconditioning (estimate = -0.805, SE = 0.021, df = 12, p = 0.001), but no significant effects were found for Days Reconditioned (Appendix C). The condition of the kelts at the time of tagging and release was significantly lower than upon their arrival at the hatchery. The intercept, representing the mean condition value for the Arrival condition, was positive at 0.805 (SE = 0.137).

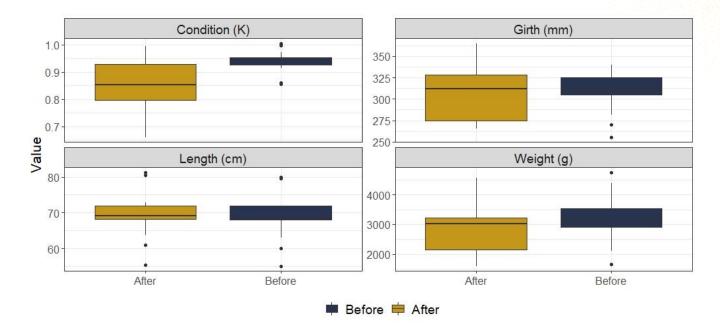
The variance component attributed to the random effect of individual fish suggested moderate variability in baseline condition among the fish, with a variance estimate of 0.002 and a standard deviation of 0.052. This indicates that while there is some individual variation among the kelts, but the treatment effect is consistent across the population. The residual variance was notably small (Variance = 0.003, S.D. = 0.053), underscoring the model's effectiveness in capturing the main effects of interest.

Our Type III Analysis of Variance further supported these findings, demonstrating a significant negative effect of condition on the measured value (F(1, 12) = 17, p = 0.001). This significant p-value corroborates the importance of hatchery reconditioning treatment in influencing the condition of steelhead kelts.



**Table 1.** Summary of 'Arrival' and 'Release' biodata for steelhead kelts captured on the Cowichan River in the spring of2021.

2021			TAG		AT CAPTURE				AT RELEASE			
PIT ID	CAPTURE DATE	RELEASE DATE	TYPE	SEX	LENGTH (MM)	WEIGHT (KG)	CIRCUMFERENCE (MM)	CONDITION (FULTON'S K FACTOR)	LENGTH (MM)	WEIGHT (KG)	CIRCUMFERENCE (MM)	CONDITION (FULTON'S K FACTOR)
989.001006677568	03-21	04-20	AC	F	685	2.98	310	0.92	687	2.14	275	0.66
989.001006677766	03-20	04-20	AC	М	795	4.76	310	0.94	813	4.58	365	0.85
989.001006677482	03-31	04-20	AC	F	630	2.14	270	0.85	638	1.98	268	0.76
989.001006677510	03-21	04-20	SAT/AC	F	715	3.34	340	0.91	718	3.02	314	0.82
989.001006677480	03-18	04-20	SAT/AC	F	720	3.54	335	0.95	728	3.08	312	0.80
989.001006677502	03-20	04-20	SAT/AC	F	680	2.96	305	0.94	692	3.2	328	0.97
989.001006677471	03-20	04-20	SAT/AC	F	701	3.28	315	0.95	719	3	302	0.81
989.001006677521	03-19	04-20	SAT/AC	F	550	1.66	255	0.99	554	1.58	265	0.93
989.001006677509	03-20	04-20	SAT/AC	F	680	3.16	325	1.00	687	3.22	319	0.99
989.001006677499	03-21	04-20	SAT/AC	F	680	2.92	310	0.92	682	2.96	311	0.93
989.001006677561	03-29	04-20	SAT/AC	F	720	3.56	321	0.95	720	3.22	337	0.86
989.001006677523	03-18	04-20	SAT/AC	F	800	4.40	340	0.86	805	3.98	331	0.76
989.001006677519	03-18	04-20	SAT/AC	F	600	2.10	282	0.97	610	2.10	275	0.93



**Figure 2.** Condition factors for Cowichan River steelhead kelts before (Arrival) and after (Release) reconditioning in the hatchery environment.

#### Post-tagging Days at Large

Average days at large (days alive post-tagging and release) across all three steelhead kelt MiniPat study years were highly variable (Table 2). Kelts from the 2021 release group were found to be inbetween the two years with shorter reconditioning times averaging 21 days at large post-tagging, with the shortest duration being 4 days and the longest 69 days (Table 2).

Table 2. Days at large for steelhead kelts post-tagging across the larger MiniPat study.

TAGGING YEAR	Ν	AVERAGE DAYS AT LARGE (MIN - MAX)
2021	10	21 (4 - 69)
2022	7	33 (5 - 74)
2023	5	11 (6 - 18)

### DISCUSSION

Results from the study suggest that for the period of treatment (~30 days), the condition factor of Cowichan steelhead kelts was not increased through reconditioning (treatment, holding, feeding, etc.). Contrary to expectations, the kelts' condition, as measured just before tagging and release, was determined through LMM analysis to be significantly lower than their condition upon arrival. Further, days at large post-tagging between the three years of the MiniPat study, demonstrated that the 1-month reconditioning trails did not likely have an overall influence on post-tagging survival (positive or negative). Additionally, none of the kelts from this study were detected returning to spawn in subsequent years. These findings suggest that while the hatchery reconditioning process is intended to improve the health and condition of steelhead kelts, there may be factors at play that result in a decreased condition.

The primary objective of this study was to increase the condition of steelhead kelts for a MiniPat satellite tag study in partnership with the Province (Davies et al. in Prep). However, given the inability to effectively increase the condition of kelts in a relatively short timeframe and the associated costs using these methods, this prolonged reconditioning study was halted after the 2021 season. In the following two years of the MiniPat tagging program, steelhead kelts were treated in the same fashion but only held for ~1 week, in the hatchery setting, before tagging and release.

The secondary objective of the study was to determine if a kelt reconditioning program could be efficient and effective in the Cowichan River. With no kelts returning from the 2021 reconditioning trials, the logistical and monetary challenges with kelt reconditioning on the Cowichan River, this method is not likely to be successful.

In other studies, kelts are often caught in downstream traps located at more permanent fishways/fences, allowing for a cost-effective and less stressful means of capture, as completed by the Columbia River Inter-Tribal Fish Commission (BPA 2016). In our study, kelts were captured using angling techniques, transferred to holding tubes, then to live wells, and then transported to the Freshwater Trout Hatchery in Duncan, B.C. The associated financial costs for capture and transport, plus the additional handling of fish post-capture, are areas of concern. Further, other kelt reconditioning programs e.g. Yakima River, have kept kelts (~1000 individuals annually) for up to 10 months before release, and they have observed increases in length, weight and condition factor, although on average 62% of the kelts perish in holding prior to release (Hatch et al 2013). Our study scope was small (N = 20), as this was a pilot program. However, VITH does not have the capacity to conduct a larger program that may be required to achieve meaningful results.

While kelt reconditioning has been moderately successful in the Columbia and Yakima rivers, these locations had the benefits of hardened infrastructure, which allowed for the easy capture of a large number of kelts. Further without fishways and fences in place on the Cowichan, increasing returns in a meaningful way through kelt reconditioning is unlikely to be successful. Results suggest that kelt

reconditioning is not an effective means of increasing steelhead abundance at the scale of this project. Costs to capture, transport, hold, and feed steelhead kelts are inhibitory and logistically challenging.

#### **Study Limitations**

The limitations of this study are recognized, particularly regarding the small sample size (N = 13) and lack of a control group to measure the direct effects of reconditioning. A control group is logistically challenging as we cannot monitor the health and condition of the same individual steelhead kelts in the river over time. However, future attempts at reconditioning could continue to sample steelhead kelts from the river to provide a baseline for changes in condition relative to the treatment group. The results derived from the linear mixed model may be biased; however, the large negative effect size of the treatment indicates a significant decline in condition despite these limitations. Each kelt had to be treated as an individual, which constrained our ability to analyze all potential variables that could influence their condition during treatment.

Consequently, while the findings highlight critical insights, the small sample size and potential biases suggest the need for caution when generalizing these results. However, this result does align with previous work (Hatch et al. 2013).

#### Recommendations

Methods to potentially increase the effectiveness of a larger kelt reconditioning program, to increase rates of iteroparity, may include:

- 1. Increase the number of kelts reconditioned. Installing a downstream capture facility on the Cowichan River during kelt migration timing (March-May). This, however, will remain a costly and high-risk process.
- 2. Increase reconditioning duration. The short duration was chosen due to the goals for the tagging study. However, if a larger program is to be undertaken a much longer reconditioning time (> 30 days) will need to be achieved. Other studies suggest that longer holding times may be more successful (Hatch et al. 2013).
- 3. Control group, as described above.



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# **APPENDIX A: PHOTO PLATES**



Photo 1. Steelhead kelt transport in 600 l tank, April 20, 2021.



Photo 2. Bio sampling steelhead kelts at the FFSBC hatchery site at Boys Road, April 20, 2021.



**Photo 3**. Netting reconditioned fish out of the 5 m round tank at the FFSBC hatchery site on Boys Road, April 20, 2021.



Photo 4. Steelhead kelt with PSAT tag and 'back pack' prior to release, April 20, 2021.



Photo 5. Releasing reconditioned Steelhead kelt in to the lower Cowichan River, April 20, 2021.



Photo 6. Releasing reconditioned steelhead kelt in the ocean, near Satellite Channel, April 20, 2021.

# **APPENDIX B: BIODATA**

DATE	PIT TAG ID	SEX	LENGTH AT CAPTURE (M)	WEIGHT AT CAPTURE (G)	CIRCUMFERENCE AT CAPTURE (MM)	РНОТО #	DATE OF MORTALITY
18-Mar-21	989.001006677480	F	720	3.54	335	1931	N/A
18-Mar-21	989.001006677523	F	800	4.4	340	1933	N/A
18-Mar-21	989.001006677519	F	600	2.1	282	1934	N/A
19-Mar-21	989.001006677521	F	550	1.66	255	1935	N/A
20-Mar-21	989.001006677476	М	795	4.76	370	1936	N/A
20-Mar-21	989.001006677471	F	701	3.28	315	1937	N/A
20-Mar-21	989.001006677508	М	555	1.58	255	1938	26-Mar-21
20-Mar-21	989.001006677509	F	680	3.16	325	1939	N/A
20-Mar-21	989.001006677502	F	680	2.96	305	1940	N/A
20-Mar-21	989.001006677531	F	760	4.1	335	1941	23-Mar-21
21-Mar-21	989.001006677510	F	715	3.34	340	1942	N/A
21-Mar-21	989.001006677503	F	710	3.64	340	1943	25-Mar-21
21-Mar-21	989.001006677568	F	685	2.98	310	1944	N/A
21-Mar-21	989.001006677499	F	680	2.92	310	1945	N/A
22-Mar-21	989.001006677538	М	630	2.48	335	1946	23-Mar-21
22-Mar-21	989.001006677554	F	700	2.84	300	1947/1948	18-Apr-21
29-Mar-21	989.001006677561	F	720	3.56	321	1949	N/A
31-Mar-21	989.001006677482	F	630	2.14	270	1950	N/A
01-Apr-21	989.001006677541	F	755	4.24	345	1951	03-Apr-21

 Table 1. Summary of all Kelt data post-capture with date of mortality.

SAT TAG SERIAL NUMBER	PIT TAG ID	ACOUSTIC TAG	TAG TYPE	DEPLOYMENT START DATE/TIME	DEPLOYMENT STOP DATETIME	FORK LENGTH (MM)	WEIGHT (KG)	SEX
20P2663	989.001006677521	47516	S/A/P	2021-04-20 15:45	05/15/2021	554	1.58	F
20P2666	989.001006677502	47523	S/A/P	2021-04-20 15:45	04/25/2021	692	3.2	F
20P2667	989.001006677561	47520	S/A/P	2021-04-20 15:45	04/28/2021	720	3.22	F
20P2668	989.001006677523	47322	S/A/P	2021-04-20 15:45	06/28/2021	805	3.98	F
20P2669	989.001006677509	47521	S/A/P	2021-04-20 15:45	04/28/2021	687	3.22	F
20P2670	989.001006677510	47518	S/A/P	2021-04-20 15:45	04/25/2021	718	3.02	F
20P2683	989.001006677471	47517	S/A/P	2021-04-20 15:45	06/17/2021	718	3	F
20P2684	989.001006677499	47515	S/A/P	2021-04-20 15:45	04/24/2021	682	2.96	F
20P2685	989.001006677480	47519	S/A/P	2021-04-20 15:45	04/26/2021	728	3.08	F
20P2686	989.001006677519	47507	S/A/P	2021-04-20 15:45	05/12/2021	610	2.1	F
N/A	989.001006677568	47512	A/P	2021-04-20 15:45	N/A	687	2.14	F
N/A	989.001006677476	47514	A/P	2021-04-20 15:45	N/A	813	4.58	Μ
N/A	989.001006677482	47513	A/P	2021-04-20 15:45	N/A	638	1.98	F

 Table 2. Summary of kelt tag and release data for Cowichan River.

#### **APPENDIX C: LINEAR MIXED MODEL AND ANOVA RESULTS**

Linear mixed model fit by REML. t-tests use Satterthwaite's method ['lmerModLmerTes t'] Formula: value ~ condition + days\_reconditioned + (1 | fish)
Data: long\_df REML criterion at convergence: -46.6 Scaled residuals: Median Min 1Q Мах 3Q -2.343062512977 -0.492585977453 -0.074482105930 0.393798068647 1.385653678821 Random effects: Groups Name Variance Std.Dev. (Intercept) 0.00272381366844 0.0521901683121 fish 0.00285287084629 0.0534122724314 Residual Number of obs: 26, groups: fish, 13 Fixed effects: df t value Estimate Std. Error Ρ r(>|t|) (Intercept) 0.80560730149829 0.13775271891187 11.12797980606095 5.84821 0 .00010619 \*\*\* 0.02095001687639 12.00000000548098 -4.16690 0 conditionTag -0.08729667241088.00130663 \*\* 0.00457474931974 11.00000001246719 0.97713 0 days\_reconditioned 0.00447014136813 .34950337 Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 Correlation of Fixed Effects: (Intr) cndtnT conditionTg -0.076 dys\_rcndtnd -0.989 0.000 Type III Analysis of Variance Table with Satterthwaite's method DenDF F value Sum Sq Mean Sq NumDF Pr(>F) condition 0.04953460859108 0.04953460859108 1 12.0000000548 17.36307 0.0013066 \*\* days\_reconditioned 0.00272389287873 0.00272389287873 1 11.0000001247 0.95479 0.3495034

Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1





#### **BRITISH COLUMBIA CONSERVATION FOUNDATION**



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