Population demographics and diet content analysis of a resident population of Coastal Cutthroat Trout (*Oncorhynchus clarkii clarkii)* in upper Shelly Creek, Parksville, British Columbia

An undergraduate research project by

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ABSTRACT

Riparian vegetation plays an integral role in the contribution of nutrients and food items to oligotrophic stream ecosystems. Thus, riparian management is a significant objective of stewardship groups interested in the conservation of aquatic fauna such as the coastal cutthroat trout (*Oncorhynchus clarkii clarkii*). The Mid-Vancouver Island Habitat Enhancement Society has recognized Shelly Creek as one of the last salmonid-bearing tributaries within the Englishman River watershed in Parksville, British Columbia. The uppermost reach of Shelly Creek is home to resident cutthroat trout which are of high conservation priority due to in-stream habitat degradation. The objective of this study was to determine the abundance of cutthroat trout within upper Shelly Creek and their diet composition. Seventy-five trout were captured by pole seine in August, September and October, 2018. Each trout was weighed, measured, clipped and sampled for stomach contents via gastric lavage. Using the Schnabel mark-recapture methodology, the population was estimated to contain 135 trout with a 95% confidence interval between 85 and 225 individuals. Diet contents indicated that aquatic and terrestrial prey items were present in similar abundance and diversity among all age classes and across all three sampling periods. Fish condition factor (calculated from fork length and mass) was significantly lower than what is expected for healthy salmonids and thus implies competition or food limitation may be occurring. These results suggest that riparian habitats contribute food items significantly to small, resident trout populations diets during low-flow conditions.

INTRODUCTION

Riparian habitats adjacent to lotic waterways provide critical ecological services such as regulating water temperature, providing allochthonous energy and promoting community diversity (Chan *et al*., 2008; González *et al.*, 2017). As a result, many aquatic organisms have unique physiological and ecological requirements that directly depend on the quality and diversity of adjoining riparian communities (González *et al.*, 2017). In efforts to conserve these interactions and the species residing within these freshwater communities, riparian areas have been recognized as critically significant habitats that require both preservation and restoration in order to maintain freshwater quality and the resilience of aquatic ecosystems (Richardson *et al.,* 2010).

One factor considered in classifying riparian areas as critically significant habitats is their disproportionate contribution to the habitats of economically and ecologically significant fishes. Salmonids are a taxon of vital fisheries importance that are particularly sensitive to the disturbance and degradation of riparian habitats (Ryan and Kelly-Quinn, 2015; Studinski *et al.*, 2017). One such salmonid species is the coastal cutthroat trout (*Oncorhynchus clarkii clarkii*), a blue-listed species of special concern in British Columbia (Species at Risk, BC, 2009) with a noteworthy dependency on small streams lined with robust riparian communities (Hilderbrand, 2003; Huusko *et al*., 2007). Current population estimates of cutthroat trout are a fraction of historical levels, with the driving factors of decline being primarily habitat or water quality degradation and low survival rates of young-of-the-year and sub-adult trout (Hilderbrand, 2003). Despite these challenges being continuously exacerbated by anthropogenic activities (Williams *et al.*, 2009), the conservation of existing riparian habitats functions to mitigate some of the concerns by providing cooler waters, feeding opportunities and protection from predators (Ryan and Kelly-Quinn, 2015; González *et al.*, 2017).

During the summer and autumn months, cutthroat trout occupying small streams (~2 m channel width) with coniferous riparian areas predominantly forage on terrestrial invertebrates that have fallen from riparian vegetation and into the water (Li *et al.*, 2016). Terrestrial-based foraging is significant as, during these months, the stream discharge is typically at its lowest for the year and, consequently, aquatic invertebrate communities in the benthos and drift are either significantly depleted or entirely nonexistent (Nicolas *et al.,* 2005). As a result, foraging options are limited and may primarily consist of terrestrial invertebrates that have fallen into the stream (Chan *et al.,* 2008). Additionally, mean water temperatures in these streams are approximately maximal during this summer-to-autumn transition. Consequently, fish metabolic rates increase with warming waters and thus adequate prey availability is required to ensure that the metabolic requirements of the population are met (Hammock and Johnson, 2014; Li *et al.,* 2016). It is critical that the energetic demands of fish are met such that they are capable of reproducing. The longevity of these resident fish populations depends not only on the reproductive success and survival of mature fish, but also the proportion of viable offspring. To promote the survival of juvenile and sub-adult fish, it is important that they are able to sequester energy into biomass to provide optimal body mass for both overwinter survival and future spawning success (Huusko *et al*., 2007; Osterback *et al.*, 2014).

Resident fish populations are defined as residing in an isolated watercourse and are unable to emigrate (or receive immigrants) due to significant barriers such as impassable waterfalls. Resident subpopulations of coastal cutthroat trout are of particularly high risk of extirpation due to their long-term genetic isolation and gradual adaptation to their environment.

The lack of genetic diversity possessed by these populations underlies their inherent susceptibility to disturbances in habitat quality (Whiteley *et al.*, 2010). Consequently, these populations may be limited in their ability to adapt to the changes in water temperature and food availability that may be the result of climate change. To assist these populations with adapting to climate change additional conservation efforts, such as riparian conservation, may be required to protect the viability of resident trout populations (Williams *et al.*, 2009) .

In addition to a lack of genetic-based adaptability, many of these resident fishes occupy streams that have been heavily influenced by urbanization within their respective watersheds. Along with a reduced inherent ability to adapt to change, these fish are exposed to habitat-related challenges including excess nutrient loading, sediment deposition and bioaccumulation of chemical contaminants (Paul and Meyer, 2008; González *et al.*, 2017). These factors more easily degrade the water and habitat quality of small streams due to their relatively low annual discharge rate (Paul and Meyer, 2008). The degradation of water quality and in-stream habitat quality may impede the ability of aquatic invertebrates to reproduce and thus feed predatory trout (Richardson *et al.*, 2010). As a result, trout residing in streams with water quality issues more heavily rely on prey items from terrestrial sources (Kraus *et al.*, 2016). The impact of water and habitat quality degradation is inherently more pronounced within small streams due to their small catchment size and relatively weak discharge rates (Kraus *et al.*, 2016). However, a disproportionate number of cutthroat trout reside in these small, perennial streams as rearing and overwintering habitat (Huusko *et al.*, 2007). Despite their value as fish rearing habitat, the narrow channel width of small streams typically results in underrepresentation in maps guiding land use practices. As a result, many of these small, trout-rich streams (and the riparian habitat associated with them) are under-protected by current legislation and environmental practices (Richardson *et al.*, 2010; Rosenfeld *et al.*, 2002).

The Englishman River watershed is situated on the southwestern coast of Vancouver Island in Parksville, British Columbia (Figure 1). Within the Englishman River watershed, most small tributaries are absent of salmonid populations with the exception of Shelly Creek. In the uppermost reach of Shelly Creek, there is a small population of resident coastal cutthroat trout that has been identified as being at high risk of extirpation due to small population size, degraded habitat quality and residency within an urban watershed (Law *et al*., 2016). These factors result in a population that is particularly vulnerable to future habitat or climate disturbances. The Mid-Vancouver Island Habitat Enhancement Society (MVIHES) has been involved in the habitat protection and restoration of many Vancouver Island watersheds and has recently prioritized the conservation of the resident trout population in Shelly Creek.

This study aims to estimate the abundance of trout within the upper reach of Shelly Creek and to determine the proportions of age-classes within the population. Additionally, this study assesses the proportion of prey items consumed that originate from terrestrial or aquatic habitats. Specifically, I hypothesized that the riparian habitat contributes a significantly greater proportion of prey items to the diet content of the fish than other aquatic subsidies during low-flow conditions. I anticipated that as a result of low-flow conditions, invertebrate prey items that would normally be in the aquatic drift would be absent or infrequently available for trout to predate upon; whereas terrestrial organisms that had fallen into the standing pools would offer a greater contribution of prey items. This hypothesis was tested by collecting diet samples from the Shelly Creek trout population during low-flow conditions and identifying organisms to the Order and origin (terrestrial or aquatic). Results may elucidate the riparian contribution to the diet of resident salmonid populations during low-flow conditions.

MATERIALS AND METHODS

Study Site

This study took place within upper Shelly Creek, Parksville, British Columbia. Shelly Creek is a small, groundwater-fed creek that originates at the base of Little Mountain and flows for nearly 6.5 km, draining into the Englishman River approximately 2.6 km upstream from the Strait of Georgia (Hilsen and Hill, 2014). The Hamilton Road Park is transected by approximately 500 m of Shelly Creek, roughly 100 m of which is the study area for this project (Figure 1). The study area is situated approximately 1.7 km upstream from the Shelly Creek and Englishman River confluence. Within this reach, a small population of resident cutthroat trout has been identified as being a significant conservation priority due to the degraded habitat, small population size and risk of exposure to future disturbance (Law *et al.,* 2016). Downstream from the study area, a series of suspended culverts and high-gradient glides prevent the trout population from emigrating or receiving immigrant, migratory trout. The reach of Shelly Creek downstream of the study area is also heavily channelized and passes through several agricultural fields with limited riparian vegetation.

The riparian area surrounding this reach of Shelly Creek is part of the Coastal Douglas Fir (CDF) biogeoclimatic zone and thus the riparian community is dominated by Douglas Fir (*Pseudotsuga menziessi*), Western Red Cedar (*Thuja plicata*) and Bigleaf Maple (*Acer macrophyllum*) (Egan and Fergusson, 1999; Law *et al.*, 2016). The vegetative community within the riparian habitat offers a closed canopy over the studied portion of the creek. Within the study site, the channel width of Shelly Creek does not exceed 3 m and the channel profile has a mean gradient of approximately 3% (Law *et al.,* 2016). During the late summer and early autumn months, there is minimal flow in this reach and thus fish are primarily isolated in small pools of a maximum of 1 m depth. Across sampling dates mean water temperature in these pools remained relatively constant at approximately 14 ± 1^0C.

Fish Sampling

Cutthroat trout were sampled on August 16th, September 13th and October 21st, 2018. Sampling began approximately 2 hours after sunrise on each date to facilitate comparison between sampling events. Additionally, sampling was conducted in the morning to optimize capture probability as juvenile salmonids are crepuscular feeders that exhibit their highest daily activity levels at dawn and dusk (Metcalfe *et al.*, 1999). On each sampling date, each of 8 pools in the study area were sampled three times with a 2-person, 5-mm mesh pole-seine (method standardized by the Kentucky Division of Water (2010)). Fish were transferred to a bucket equipped with oxygen bubblers, and water temperature and dissolved oxygen levels were monitored using a Polaris OxyGuard® electronic probe. Fish were individually anaesthetized in a solution of buffered Tricaine Methanesulfonate (TMS™) at approximately 75 mg/L, until the fish had reached stage II anaesthesia (partial loss of equilibrium). Once anaesthetized, individual trout were weighed to the nearest 0.1 g and fork length (FL) was measured to the nearest mm. For fish that were captured for the first time, the adipose fin was removed with scissors. Both newly captured (adipose fin present) and recaptured (adipose fin absent) fish were recorded and used for estimating the population size. Captured fish were observed to occur in three distinct, non-overlapping size classes, these groupings were interpreted as representing age-0 (<90 mm FL), age-1 (90-160 mm FL) or age-2+ (>160 mm FL). All fish were also examined for ectoparasites although none were examined during this study.

Gastric Lavage

Gastric lavage is a common, non-lethal and efficient method of sampling diet among salmonids of a variety of age-classes (Kowalik, 2016; Studinski *et al.*, 2017). Stomach contents were removed via gastric lavage, in which a small, blunt plastic tube attached to a syringe was gently inserted through the mouth to the posterior end of the stomach (when the tubing can no longer advance). By compressing the syringe to apply a constant flow of water and gradually retrieving the tubing, diet contents were flushed from the gut and were collected through the mouth into a funnel adapted with a 100-µm mesh sieve. Each stomach was flushed twice, and the procedure was consistently completed in less than 30 seconds. Stomach contents were stored in a solution of 95% ethanol. Fish were then transferred to an aerated recovery bucket, monitored until fully recovered and released back to the same pool in which they had been captured.

Additionally, the VIU Department of Fisheries and Aquaculture donated three juvenile, captive-bred rainbow trout (*Oncorhynchus mykiss*) to use in testing the effectiveness of the gastric lavage technique and apparatus. Once processed, these fish were euthanized and frozen. Of the fish sampled in Shelly Creek, one age-0 cutthroat trout did not recover from the anaesthetic and was subsequently euthanized and frozen. All four of these fish were later thawed and dissected to assess the effectiveness of the gastric lavage methodology. Although the rainbow trout were fed a commercial pellet-based diet, dissections revealed that gastric lavage was effective at removing all food items from the stomach. These results were similarly observed in the wild cutthroat trout mortality, indicating that the apparatus and method were appropriate to effectively retrieve stomach contents.

Stomach Contents

Due to the small sample size of age-2+ trout (n = 5), diet content analysis only pertains to age-0 and age-1 trout. Diet contents were identified and counted in the laboratory under a dissecting microscope and prey items were identified to Order using a dichotomous key (Merritt and Cummins, 1995). Prey items were further categorized as terrestrial or aquatic origin (as outlined in Appendix A1). For example, larvae and nymphs of aquatic taxa were classified as aquatic, while winged adults were classified as terrestrial. Prey items of a strictly aquatic life history stage were classified as *aquatic* for the purposes of this study. In contrast, prey items from the air, soil or vegetation were categorized as being *terrestrial*. Furthermore, small (<2 mm) nematodes were found in 40.0% (30 out of 75 samples) of the trout diet samples in an aggregated distribution (low intensity infections were common, and high intensity infections were uncommon). As a result of their small size relative to other prey items and aggregated distribution these nematodes were assumed to be parasitic and not prey items; however, this assumption could not be fully validated without a higher resolution taxonomic identification.

Data Analysis

The proportion of recaptured fish to newly captured fish (as indicated by the presence or absence of an adipose fin) was used to estimate the population size within the study area. Population size was estimated using the Schnabel capture-mark-recapture methodology as depicted in Equation 1, with a 95% confidence interval calculated by Equation 2. As the summed count of recaptures was less than 50 individuals, Krebs (1999) suggests using values from a Poisson distribution as the denominator of Equation 2, with x being equal to the summed number of recaptures.

Equation 1. Schnabel capture-mark-recapture method in which $\widehat{N}$ is the mean estimate of the population size, C_t is the number of captured animals, M_t is the total number of marked animals, and R_t is the number of recaptured animals at a specific sampling session in which *t* is the time of the final sampling effort.

$$\widehat{N} = {\Sigma\,(C_t\ \cdot M_t)}\Big/{(\Sigma R_t)}$$

Equation 2. Lower and upper limits of 95% confidence interval of population size estimated by the Schnabel capture-mark-recapture method. With 16 recaptured individuals, denominator values were obtained from Table 2.1 in Krebs (1999).

$$Confidence\ Limit\ (upper\ or\ lower) = \frac{\Sigma\,(C\ \cdot M)}{x} = \#\ \text{of individuals}$$

Prey item enumeration yielded count values for each taxon consumed by individual fish, and within the population at each sampling period. Diet diversity was quantified using the Simpson's (1 - D) Index of Diversity (Krebs, 1999). Diet diversity was calculated for individual trout and the average value of all fish sampled within a sampling date was compared between sampling periods using a Kruskal-Wallis one-way ANOVA. To investigate if trout differentially foraged on terrestrial or aquatic organisms, the counts of each prey source were compared between age classes and sampling dates. Terrestrial and aquatic prey counts were paired within stomach content samples and had non-normal distributions, the groups were compared using the Wilcoxon signed-rank test (Zar, 2010).

The fork length and body mass of each individual trout was recorded, and results are summarized in Table 2. These metrics were utilized to calculate Fulton's Condition Factor (Equation 1) in order to estimate the relative condition factor of the trout (Table 2) (Barnham and

Baxter, 1998; Froese, 2006). Condition factor was non-normally distributed among age classes and sampling dates as indicated by the Wilks-Shapiro test for normality. Condition factor was compared between the three sampling dates using a Kruskal-Wallis one-way ANOVA and grouped data from both age-0 and age-1 trout. Age-0 and age-1 trout data were grouped together as condition factor was similar between these groups across all sampling dates (tested with three Mann-Whitney *U*-tests). Fork length and body mass comparisons were conducted for age-0 and age-1 trout using a Kruskal-Wallis one-way ANOVA. Pair-wise comparisons were conducted with a Mann-Whitney *U*-test. Among salmonids, a condition factor significantly below 1.0 is indicative of poor overall condition and a disproportionately long body relative to their mass (Barnham and Baxter, 1998). Mean condition factor was compared to the value of 1.0 using a one-sample Mann-Whitney U-test. All statistical comparisons were conducted using the *Real-stats* Microsoft Excel® add-in at a confidence level of 95% ($\alpha = 0.05$).

Equation 3. Fulton's Condition Factor in which $N = 5$, W = fish wet weight in grams, L = fork length in mm.

$$Condition\ Factor\ (K) = \frac{10^N\ W}{L^3}$$

Animal Care and Permitting

All research procedures were approved by the Vancouver Island University Animal Care Committee (VIU ACC) under the Animal Use Protocol (AUP) # 2018-03-R-DEMERS. Additionally, fish capture and sampling were permitted by the British Columbia Ministry of Forests, Lands and Natural Resource Operations under permit NA18-358355.

RESULTS

Population Size and Structure

Among the three sampling periods, within the upper reach of Shelly Creek, a total of 83 fish were captured (68 individuals and 16 recaptures) (Table 1). Individuals were predominantly age-0 (n = 35 (52.9%)), age-1 and age-2+ (n = 27 (39.7%) and n = 5 (7.4%), respectively). The recapture rate among age-0 trout was the lowest, with only two fish recaptured out of the 36 individuals handled. In contrast, age-2+ trout were most frequently recaptured, as four out of five handled fish were recaptures (Table 1). The Schnabel Mark-recapture methodology suggests a population size (of all sizes) of 135 trout with a 95% confidence interval ranging from 85 to 225 individuals.

Trout Mass, Length and Condition Factor

Wet weight, fork length and condition factor were compared within age classes and between sampling periods (Figure 2). The wet weight of age-0 and age-1 fish did not vary significantly between the three sampling dates (H = 7.8; df = 2; P = 0.194 and H = 2.59; df = 2; P = 0.275 respectively). However, the fork length of age-0 trout increased significantly between sampling periods (H = 18.1; df = 2; P < 0.001) (Figure 2A), whereas the fork length of age-1 trout did not vary significantly between sampling periods (H = 1.9; df = 2; P = 0.370). Within the upper reach of Shelly Creek, the condition factor of age-0 trout did not vary significantly between sampling dates (H = 4.1; df = 2; P = 0.126) (Figure 2E). In contrast, the mean condition factor of age-1 trout differed significantly across all sampling dates (U = 22; P = 0.007). The condition factor of both age-0 and age-1 trout was significantly lower than 1.0 across all sampling dates, except for age-0 fish sampled on August 16th (Table 2).

Diet Contents

Among age-0 and age-1 trout, Dipterans were the most abundant prey taxa by count across all three sampling dates (37.7 ± 0.7% SE) (Figure 3). Dipterans were mostly aquatic, larval forms (59.5%), with larger terrestrial adults comprising 40.5% of the dipteran prey items. The next most abundant prey taxa across all sampling dates was Hymenoptera (9.4 ± 1.0% SE), and by Collembola (9.3 ± 2.6% SE) (Figure 3). Noteworthy terrestrial prey taxa included a large (~30 mm) terrestrial worms (Annelida), six intact spiders (<8 mm) (Araneae), 28 barklice (Psocoptera), and 49 ants (Hymenoptera). Aside from small larval dipterans, aquatic prey items were primarily the nymphs of stoneflies (22), caddisflies (29) and mayflies (20). Results from the Wilcoxon paired-rank test indicate that there was no significant difference between the number of terrestrial and aquatic prey items consumed among age-0 and age-1 trout across all three sampling dates (Table 3; Figure 4). Additionally, the mean diet diversity (Simpson's Index of age-0 and age-1 trout did not vary significantly between sampling dates (H = 0.036, df = 2, P = 0.982) (Table 3). Two age-2+ trout were sampled for diet contents on August 16th and revealed partially digested fish tissues, thus suggesting that the larger age class of trout within Shelly Creek cannibalizes smaller fish.

DISCUSSION

Riparian habitats are associated with the regulation of stream hydrology and chemistry, while contributing indirectly to fish habitat and the trophic pathways predatory fish ultimately rely upon (González *et al.*, 2017). The results of this study further emphasize the importance of riparian habitats by demonstrating that, during low-flow conditions, the stomach contents of cutthroat trout contain a significant number of terrestrial prey items. This outcome may support that the proportion of terrestrial prey items is inversely associated with water flow among small streams.

Population Size and Structure

For this study, coastal cutthroat trout were captured during the late summer and early autumn during low-flow conditions in which fish were residing in small, perennial pools with virtually no connectivity between adjacent pools. These conditions restricted the movement of fish, and thus provided a closed population within the study area, allowing for a Schnabel mark-recapture estimate of the population size. The Schnabel mark-recapture methodology yielded a mean estimate of 135 (95% CI: 85 to 225) individuals within the 100 m study area of Shelly Creek. Rosenfeld *et al.* (2002) studied numerous small (<2 m wetted width), coastal, oligotrophic streams during the late summer months and obtained a mean estimate of 0.8 cutthroat trout per metre. Rosenfeld's estimate of cutthroat trout density is lower than the estimate obtained from upper Shelly Creek (0.85 to 2.25 cutthroat per metre). However, it is noteworthy that during these low-flow conditions much of the upstream habitat was dry and inaccessible to fish (B. Judson, pers. obs.), thus the density of fish within the study area may be seasonally inflated and non-representative of year-round habitat occupation. Additionally, Rosenfeld's research primarily focussed on anadromous cutthroat trout populations, whereas the fish within upper Shelly Creek form a resident population. There is currently little information in the literature comparing population densities between anadromous and resident cutthroat trout populations. Furthermore, the mean Schnabel mark-recapture estimate assumes that all three age classes of trout (age-0, age-1 and age-2+) have equal probability of capture. However, when the population size is estimated for each age class the sum of these estimates is more than twice that of the original, grouped estimate. This inflated estimate is likely the result of the low number of age-0 recapture events. Also, post-sampling mortality, predation events or low fish capture efficiency may result in an inaccurately inflated population size estimate. For instance, the diet contents of two age-2+ trout (both from August 16th) contained partially digested fish tissues and thus suggest probable cannibalism as no other fish species were observed in Shelly Creek. Similarly, among brown trout (*Salmo trutta*), adult trout cannibalizing juveniles was noted to be a significant contributor to juvenile mortality in freshwater habitats (Vik *et al.*, 2001). The grouped and age-specific Schnabel mark-recapture methods vary, yet both agree that the population of cutthroat trout within upper Shelly Creek is a small population. MVIHES volunteers have observed this population for years and have estimated the population to contain no more than 200 individuals; an approximation that is generally supported by the results obtained in this study.

Trout Length, Mass and Condition Factor

The fork length (FL) of age-0 trout increased between all three sampling periods, yet their body mass increased at a slower, yet non-significant rate such that the condition factor of this age-class remained relatively constant (Figure 2a, c and e). This observation has significant implications for trout recruitment, as trout of a greater mass and FL typically demonstrate greater overwinter survival and spring spawning success (Huusko *et al.*, 2007). It is also important to note that salmonid growth rates are primarily influenced by food consumption and less drastically influenced by abiotic factors such as temperature and water oxygenation (Petty *et al.*, 2014). Additionally, the mean water temperature within the study area fluctuated minimally across sampling events (due to the groundwater origin of this reach of Shelly Creek) and thus the lengthwise growth observed among age-0 trout in Shelly Creek is unlikely to be the result of thermal fluctuations. However, among salmonids, a CF of 1.0 is the reference value for a fish of a healthy mass relative to its FL (Barnham and Baxter, 1998). The trout within Shelly Creek exhibit a mean CF value significantly less than 1.0 across all age classes and sampling dates (apart from age-0 trout on August 16th) (Table 2). These results suggest that this trout population may be in sub-optimal physical health which may be the result of resource limitations.

Diet Contents

There are several studies that suggest the diet of stream-dwelling salmonids is dominated by terrestrial invertebrates that have fallen into the stream during low-flow conditions (Ryan and Kelly-Quinn, 2015; Li *et al.*, 2016; Sweka and Hartman, 2008). In contrast, I observed that trout consumed a similar proportion of prey from both terrestrial and aquatic origins (Figure 4). This observation may be the result of the groundwater contribution to Shelly Creek, which may allow the habitat to be occupied by aquatic organisms throughout the summer. However, many of the aquatic prey organisms in diet samples were small and abundant (e.g. dipteran larvae and copepods), whereas a large proportion of terrestrial prey items were infrequent but much larger (e.g. annelid worms and isopods). The disparity between prey sizes from aquatic and terrestrial habitats suggests that these habitats may differentially contribute energy to Shelly Creek. Also, the chitinous exoskeleton of many terrestrial invertebrates is durable and thus may last longer in the trout stomach, leading to an overrepresentation of terrestrial prey items (Courtwright and May, 2013). Diet content analysis revealed that the diversity and composition of prey items did not vary significantly between sampling events. This outcome is important as it suggests that the trout may not deplete the resources within their isolated habitat during low-flow conditions.

Concluding Remarks

The goal of this study was to determine the size of the coastal cutthroat trout population residing within upper Shelly Creek and to understand what this population consumes during the late summer and early autumn months. Specifically, prey items were identified with respect to their habitat of origin and the counted number of prey items from terrestrial and aquatic habitats were compared. The results of this study suggest that there is a mean number of 135 trout with a 95% confidence interval ranging from 85 to 225 individuals. Diet content analysis revealed that terrestrial and aquatic habitats contribute a similar number of prey items to the diet of cutthroat trout during low-flow conditions. Whereas riparian habitats are widely recognized as key components to maintaining ecosystem health, their intrinsic value may be further emphasized as climate changes progresses (Richardson *et al.,* 2010; Johnson and Almlöf, 2016). For instance, as freshwater systems warm, the metabolic demands of their inhabitants will increase and thus the demand for food and the resulting competition will increase (Hammock and Johnson, 2014). Additionally, under circumstances of aquatic pollution, aquatic invertebrates may be quickly extirpated, and thus the value of terrestrial food subsidies may increase dramatically (Kraus *et al.,* 2016). Thus, the conservation and restoration of riparian habitats will be vital to the integrity of freshwater ecosystems. The trout population in the uppermost reach of Shelly Creek will undoubtedly benefit from further riparian conservation as it contributes a significant proportion of dietary items to a small population with a low average condition factor. Future works may consider studying this population's diet composition and condition factor trends throughout the year or comparing diet composition between populations of cutthroat trout.

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TABLES

Table 1. Number of cutthroat trout captured and recaptured of different age classes among three sampling dates in Shelly Creek. On September 13th a single, newly captured age-0 trout died during handling. Also, it is possible that on September 13th and October 21st some recaptured fish may have been the same individual.

	Newly Captured			Recaptured			
Sampling Date	Age-0	Age-1	Age-2+	Age-0	Age-1	Age-2+	Total
August 16th	16	11	3	N/A	N/A	N/A	30
September 13th	13	7	0	0	5	2	27
October 21st	6	9	2	2	5	2	26

Table 2. Fork length, wet weight and condition factor (mean ± standard error) of cutthroat trout among three sampling dates within Shelly Creek. Mean condition factor was compared to the expected value of 1.0 using a one-way Student's t-test (Barnham and Baxter, 1998). Sample sizes provided in Table 1.

	Age	Body Mass (g)	Fork Length (mm)	Condition Factor	*P*-value
August 16	Age-0	1.0 ± 0.064	47 ± 1.0	0.886 ± 0.235	0.063
	Age-1	14.1 ± 2.4	116 ± 5.6	0.926 ± 0.089	0.020
September 13	Age-0	1.3 ± 0.10	52 ± 1.1	0.887 ± 0.117	0.005
	Age-1	13.7 ± 1.5	120 ± 4.7	0.785 ± 0.029	< 0.001
October 21	Age-0	1.4 ± 0.13	56 ± 1.4	0.785 ± 0.095	< 0.001
	Age-1	10.4 ± 1.1	108 ± 5.6	0.887 ± 0.058	< 0.001

Table 3. Trout diet content diversity (mean ± standard error) and the counted number of aquatic and terrestrial prey items among three sampling dates in Shelly Creek. Data were pooled from both age-0 and age-1 trout (sample sizes available in Table 1). Differences in the number of aquatic and terrestrial prey items were compared using a Wilcoxon signed-rank test.

Sampling Date (2018)	Simpson's Index (1 - D)	Number of Terrestrial Prey Items	Number of Aquatic Prey Items	Wilcoxon Signed-rank *Z*-score	*P*-value
August 16	0.537 ± 0.059	98	109	0.309	0.363
September 13	0.518 ± 0.053	44	61	1.571	0.061
October 21	0.472 ± 0.087	42	42	0.260	0.401

Figures

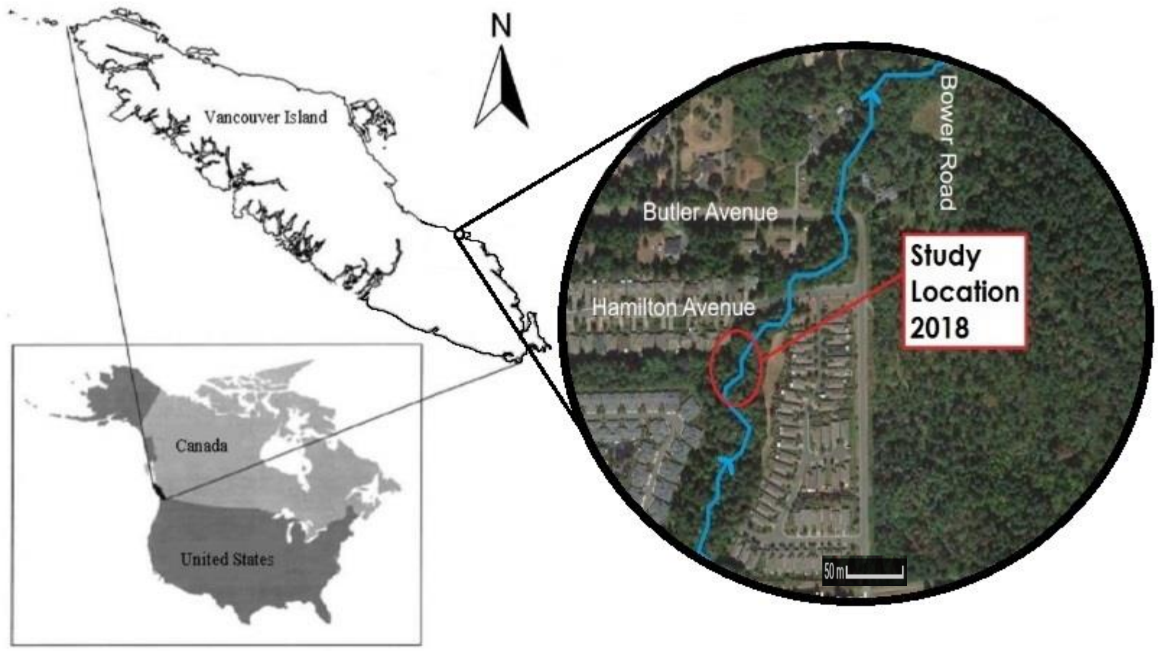

Figure 1. Location of Shelly Creek and the 2018 study location (adapted from Dumont, 2017).

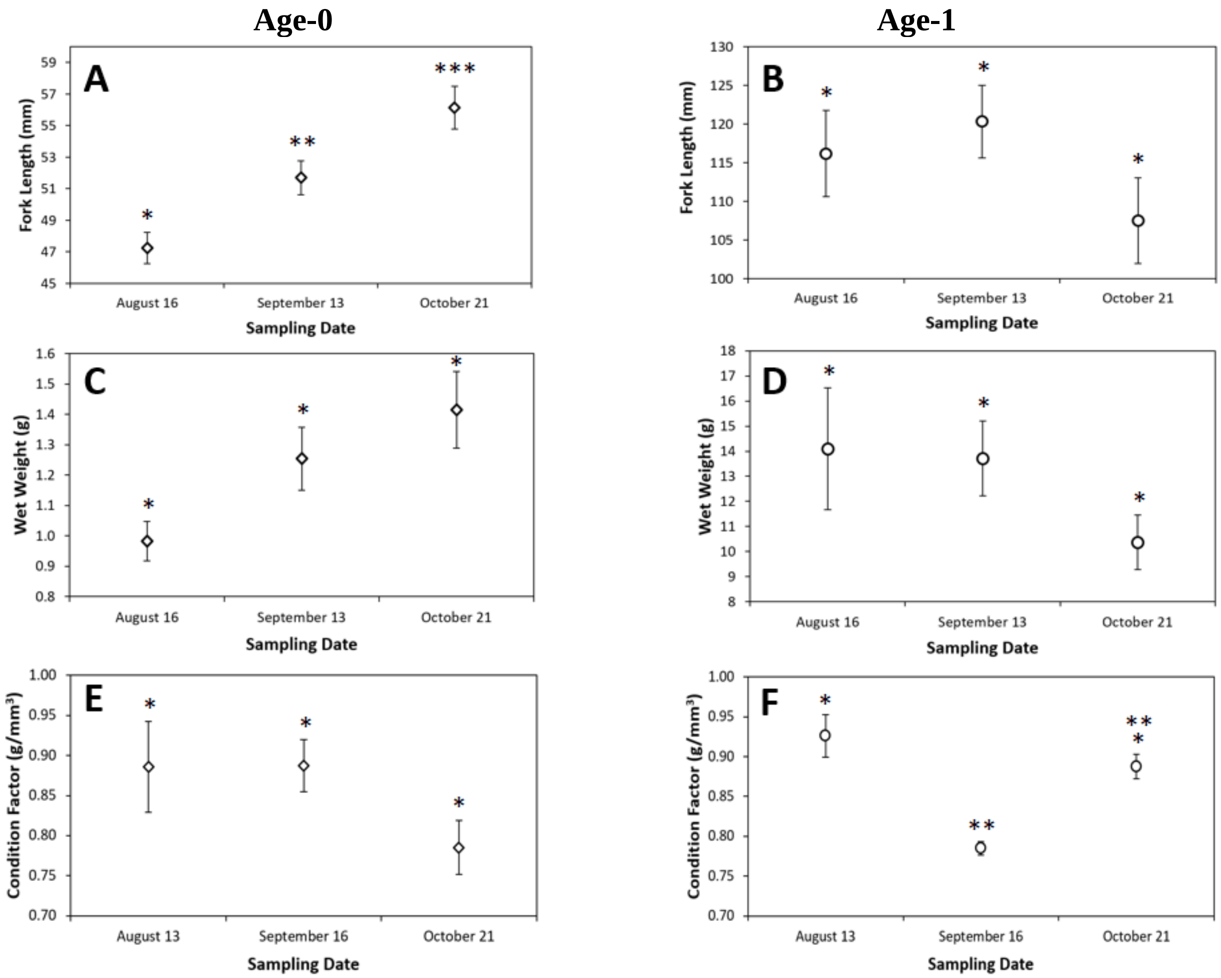

Figure 2. Mean (± standard error) fork length (A and B), wet weight (C and D) and condition factor (E and F) for age-0 (◊) and age-1 (o) cutthroat trout. Trout were sampled from upper Shelly Creek, Parksville, British Columbia across three sampling dates. Significant differences (P < 0.05) between values are indicated with different numbers of asterisks (*), whereas values with the same number of asterisks are non-significantly different from each other.

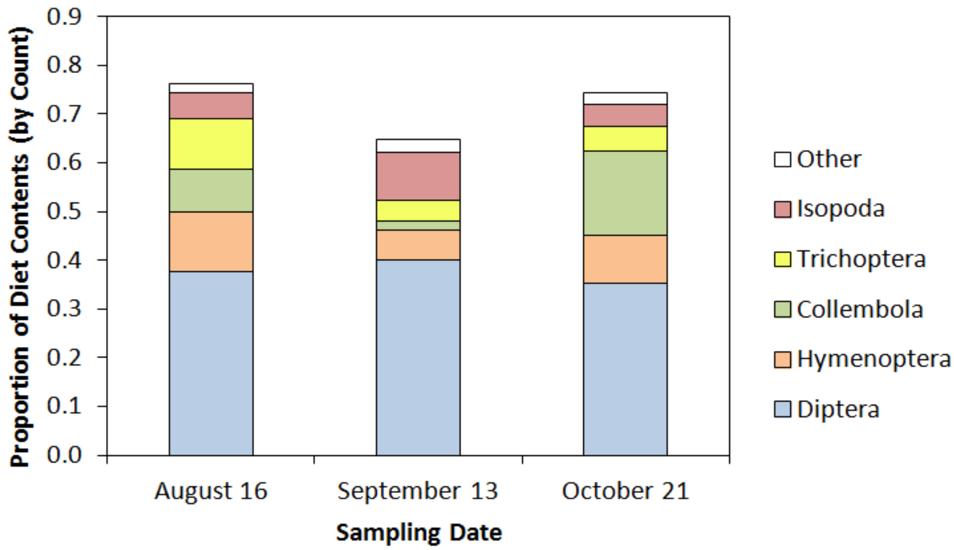

Figure 3. Mean composition of diet contents (by count) among age-0 and age-1 cutthroat trout diet samples across three sampling dates from upper Shelly Creek, Parksville, British Columbia.

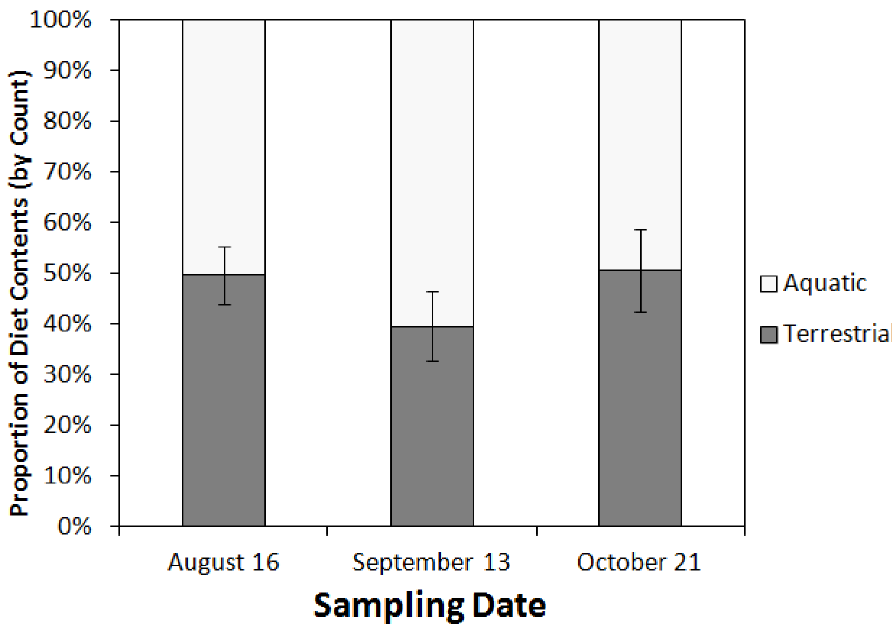

Figure 4. Proportion of prey items (by count) (mean ± SE) from both terrestrial and aquatic habitats among age-0 and age-1 cutthroat trout diet samples across three sampling dates from upper Shelly Creek, Parksville, British Columbia.

APPENDIX

Appendix A1. Prey item classification with respect to habitat origin.

Terrestrial	Aquatic
Mayflies (adults)	Mayflies (nymphs)
Caddisflies (adults)	Caddisflies (nymphs)
Stoneflies (adults)	Stoneflies (nymphs)
Dipterans (e.g. Mosquitoes) (adults)	Dipterans (larvae)
Dragonflies (adults)	Dragonflies (nymphs)
Beetles (terrestrial)	Beetles (aquatic)
Annelids worms	Annelids (aquatic)
Mites	Copepods
Springtails	
Spiders	
Hemipterans (e.g. Aphids, Leafhoppers)	
Woodlice	
Barkflies	
Crickets and Grasshoppers	
Scorpionflies	
Lacewings	
Hymenopterans (e.g. Wasps, Ants)	
Arthropods (e.g. Millipedes)	