

Gambier Island Coastal Tailed Frog Project: Field Sampling Summary and preliminary results-2022



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Introduction

The Coastal Tailed Frog (*Ascaphus truei*) project began in 2021, as a response to proposed logging in the headwaters of Whispering Creek, a first-order stream that drains a south-southeastern slope of Mt. Killam into West Bay on Chá7elkwnech/Gambier Island. There are no official government records of Coastal Tailed Frogs (CTF) presence on Gambier Island (e.g., Conservation Data Center 2021), which fails to inform professional foresters of species distributions during the development of logging plans (Forest Practices Board 2014). Consequently, plans to extract about four hectares of old forest from riparian areas in Whispering Creek headwaters were developed around the assumption that CTF are absent, even though observations by locals suggest they are present (Madrone Environmental Consultants 2009). If logging plans are carried out, then only two small unlogged patches (about 0.006 Hectares) with 20-meter-wide riparian areas will remain within a four-hectare cut block (Gambier Forest Resources Ltd. 2020). This severely threatens the survival of the CTF population, which requires extensive old forest in riparian areas (~300 meters) that sustain seasonally consistent cool and humid conditions for adult and subadult survival and dispersal, and stable cool clear stream habitats for reproductive success and tadpole rearing (Todd et al. 2014; Environment and Climate Change Canada 2016). The CTF is listed as a species of ‘Special Concern’ by the government of Canada and is protected by legislation under SARA (*Species at Risk Act*) and FRPA (*Forest Range Protection Act*). Unfortunately, the province is lacking sufficient data to properly assess the current status of the CTF. Thus, this research will contribute valuable data to fill the current gaps in knowledge, as it is crucial to have these data represented in provincial and federal databases and mapping platforms like the Species Inventory Web Explorer, Ecosystems Explorer and the B.C. Conservation Data Center. These resources inform professionals working in the resource sector, so they are aware of the distributions of SARA-listed species and facilitate adequate protection of their critical habitats.

Coastal Tailed Frogs are endemic to the Pacific Northwest, with unusual qualities among frog species such as limited hopping ability and stream-rearing tadpoles with specialized mouthparts for rearing in fast flowing water (i.e., riffles). They are absent on both Vancouver Island, as well as the Southern Gulf Islands (Environment and Climate Change Canada, 2016), which highlights the conservation significance of populations of CTF on Gambier Island. Due to the geographic isolation of Gambier Island and restricted dispersal (gene flow) with adjacent mainland populations, the CTF metapopulation(s) is both genetically unique in BC and especially vulnerable to extirpation because they are unlikely to be rescued from surrounding populations. The geographically isolated CTF populations on Gambier Island comprise unique components of biodiversity important to the long-term survival of the species and should be prioritized for conservation management actions (Nielsen et al., 2016).

Information regarding CTF populations is necessary to identify unique elements of biodiversity in BC, and together with abundance estimates, distribution data is paramount to evaluating population status, the extent of threats and determining their conservation status in BC.

(<https://www.natureserve.org/canada/who-we-are#cdcs>). Currently, the province is lacking sufficient data to properly assess their conservation status and therefore by default they are *Yellow Listed* (Apparently Secure; BC Conservation Data Center 2016). The Gambier Island CTF population data collected in this research will add critical information to inform management and protection measures that can ultimately reduce threats. A greater understanding of the distribution of Gambier Island CTF is necessary for sustainable land management, will aid in reducing riparian damage, and ensure habitat connectivity to ensemble CTF metapopulation stability. As the Atl’ka7sem/Howe Sound is now a designated UNESCO Biosphere Reserve, it is fundamental to include the protection and management of ecosystem assets to future sustainable resource management plans, biodiversity monitoring, and conservation management plans on Gambier Island.

Objectives and Hypotheses

The objective of this research is to determine the distribution of CTF on Gambier Island using environmental DNA (eDNA) sampling. We chose to use eDNA for this purpose, as it has been found to be a highly effective and cost-efficient way to determine the presence of CTF (e.g., Hobbs et al. 2019). This research is addressing the hypothesis that CTF were long isolated on Gambier Island and have therefore dispersed to all watersheds that continue to provide adequate habitats. Possible interactions with native fish (such as the Coastal Cutthroat Trout, *Ocorhynchus clarkii clarkii*) may have limited their habitat use and consequently, their distribution in some watersheds might be restricted to headwaters and above barriers that preclude the presence of fish (e.g., Heyes et al. 2006). Alternatively, the CTF has adapted to sympatric interactions with the Coast Cutthroat Trout and the species coexist together throughout their home streams. Additionally, disturbances from colonial socioeconomic activities such as settlement and industrial logging might result in range contractions or loss of populations from their home streams (e.g., Wahbe et al. 2005). For instance, we sampled in Grennon Creek, Mannion Creek, Andy's Bay Creek, New Brighton Creek, and two Whispering Creek tributaries where some degree of human disturbances to habitats might potentially limit CTF distribution and therefore, would show absence in some of our eDNA sample sites. However, despite this disturbance, we observed CTF tadpoles during our surveys this year rearing in Mannion Creek upstream of logging impacts, suggesting that there may be some resilience to some degrees of human disturbance.

Overview of Field Sampling and Methods

Our initial proposal planned for the sampling from seven of the largest watersheds on Gambier Island. We budgeted for a total of 35 sites (five sites per stream) to evaluate influences from tributaries on eDNA distribution within watersheds and included streams perceived to have different levels and types of human disturbances. Sample sites included one located near the mouth of each stream, and then additional sites distributed upstream and spaced within 1.5 km to evaluate the distribution of habitat use within each stream. Blanks consisting of distilled water filtered through the apparatus and were collected at the beginning and end of each sampling day to ensure the sampling apparatus and method were not collecting false positives (i.e., tailed frogs eDNA detected at sites where they are absent). For field planning, we plotted proposed sample locations within the creeks on maps and considered stream lengths and presence of larger tributaries. However, final sample site selection occurred in the field to include only those locations with flowing surface water, rearing habitats for tailed frog tadpoles.

During our field days, we needed to adjust our sampling plan due to elements such as accessibility of sampling sites, insufficient surface water flow, and considerations of private property. For example, some tributaries were either completely dried up (subsurface) or in standing pools, which often resulted in our collecting fewer sample sites in some streams during summer period. Consequently, we had 17 sample kits remaining at the end of summer and used those to continue sampling during October and December, resulting in a total of nine creeks sampled.

Sampling for eDNA used the instream filtration method as outlined by Carim et al. (2016), which captures eDNA from water pumped directly from the stream onto filters and has detected the presence of target aquatic animals located between 1.5 to 4.5 kilometers away in streams (Stamford et al. 2022). Sampling at each site also included rapid collection of habitat variables as described in BC Government RISC standards (e.g., Johnston and Slaney 1996). Our field headquarters was in West Bay on Gambier Island where we stored and maintained our equipment, charged batteries, and filtered distilled water for the blanks. This was a convenient location to begin and end each day, prepare our field equipment, and collect sampling blanks. The core sampling team included Sylvia and Samantha who were often lucky to be joined by an ACE monitor from the Skw̓wú7mesh Úxwumixw/Squamish Nation, and volunteers from the local community.

Creek Descriptions

McDonald Creek

The first creek we sampled was McDonald Creek. The field team consisted of Sam, Sylvia, and Mike, and we were lucky enough to be joined by [Peter Scholefield, President of the Gambier Island Conservancy](#). McDonald Creek is in the head of West Bay, so we were able to drive to the creek mouth and take our first sample there. This day was helpful to ensure Sam and Sylvia were confident and comfortable with the protocol that we would be using. Mike was able to confirm that we were following the protocol correctly and answer any initial questions we had. We hiked up the creek bed, and at times followed adjacent trails for easier access to the top of the creek. One major tributary was sampled which looked to be an ideal CTF habitat. While we had initially planned to sample tributaries, all except one were dry and consequently not inhabitable by CTF tadpoles.

Due to time constraints, we were only able to take four samples on that first sampling day. Sylvia was able to go back later in the summer to take two additional samples near the headwaters of the creek, resulting in a total of six samples from McDonald Creek which, in addition to three samples collected in 2021, will provide a good distribution of information.

One interesting observation during the sampling of McDonald Creek was multiple high-gradient cascades where stream flows were often underground. The low gradient sections upstream of these cascades also sustain Coastal Cutthroat Trout, which are thought to prey on CTF tadpoles and might limit their distribution within streams (Heyes et al. 2006). Previous year's (2021) eDNA sampling at one site amongst the cascades failed to detect CTF. For sampling this year (2022), we collected samples from more locations both downstream and upstream of the cascades. Consequently, results of our eDNA sampling might find an absence in some locations, suggesting a small population is limited by interactions with Coastal Cutthroat Trout. Alternatively, the interactions between the two species might influence adaptive changes that enable the CTF to sustain presence throughout the stream.



Figure 1: Sylvia on our second McDonald Creek sampling day.

Whispering Creek

The second sampling day was conducted by Sylvia and Sam in Whispering Creek where previous eDNA sampling in 2021 had already detected presence of CTF. We sampled again in 2022 to evaluate seasonal changes in eDNA distribution and presence in tributaries. The watershed is very significant to the community of West Bay being salmon-bearing, a drinking water source for many families, and lets out at the West Bay Public Dock. This creek runs alongside a local trail system, which allowed us to follow the trails for most of the sampling. The headwater areas of Whispering Creek lie within the boundaries of a working wood lot but remarkably, have escaped any recent disturbances from logging-associated activities. The middle reaches show signs of extensive logging activities from over a hundred years ago, which have since recovered to contain old forest (>100 years old). The absence of any logging signs in some areas in the headwaters of Whispering Creek suggests significant habitat areas have escaped logging-associated disturbances entirely (Maddison Consultants Ltd. 2021; Ecologic Consultants Ltd. 2021).

We were able to take four samples on our first day of sampling Whispering Creek. Unfortunately, the water flow was minimal at the headwaters, and one of the tributaries we planned to sample was dry. After this sampling day, we noticed a section in the creek that we accidentally skipped over, so the following day we sampled the creek at one more site. We were able to collect eDNA from five sites in Whispering Creek during summer, and then added three more sites in December 2022 to evaluate two tributaries entering the fish bearing zone downstream of a 10 meter falls.



Figure 2: Sam and our field set-up in Whispering Creek.

Mannion Creek

The field team for Mannion Creek included Sam, Sylvia, Mike, and Sk̓wx̓wú7mesh Úxwumixw/Squamish Nation ACE Monitor, Amy Baker. We picked Amy up at New Brighton, and then hiked up the trails to Mannion Creek. Mannion is the longest stream on Gambier Island with steep and difficult terrain, limited trails to follow, so we had to scramble up the creek to access sample sites. We took two samples, and at the second site, Mike spotted a Coastal Tailed Frog tadpole! It was our first sighting, which was very exciting for all of us. Pictures of the tadpole can be seen below. We collected duplicate samples at this location to evaluate the repeatability of our eDNA samples due to the confirmed presence of CTF.



Figure 3: First CTF tadpole sightings in Mannon Creek.



Figure 4: Amy Baker, Mike Stamford, and Samantha Wing accessing a difficult sample sites in Mannion Creek.

A second field day was needed to complete Mannion Creek sampling due to its terrain and size, so on our third summertime field day, we drove in through the logging roads, and then hiked to two downstream sampling locations. A total of four sites were sampled for eDNA during the summer in Mannion Creek and then a fifth site was sampled near the mouth in November 2022.

Andy's Bay Creek

Sam, Sylvia, and Mike sampled two sites in Andy's Bay Creek by driving into the woodlot via logging roads and hiking into the creek. The first sample ended up being very challenging to access and resulted in the three of us bushwhacking through dense forest on the side of a steep bluff for about an hour. We successfully collected eDNA from a very steep section of the creek. We sampled a second site upstream near the trailhead to Muskeg Lake which was very easy to access.

We couldn't access any lower points of the creek by foot or vehicle, so Sam's father Kevin Wing kindly brought us there on his boat, and we entered the creek through the mouth. This was our third and final sample from the difficult-to-access and steep Andy's Bay Creek.

Gambier Creek

Gambier Creek flows from Gambier Lake, which is centrally located on the island, to Douglas Bay which is on the opposite side of the island from West Bay. In order for us to access this creek, Kevin Wing took us there (stopping to collect from Andy's Bay Creek mouth on the way), and dropped us off at the Douglas Bay Campground. We camped one night there, and then hiked up along the creek to Gambier Lake. We followed the Gambier Lake trail and cut in to meet the creek at a couple of locations. We took

one sample downstream from the Douglas Bay Pools along Gambier Creek, and three more above, totaling four samples. This creek dried up completely before it reached Gambier Lake. We then camped a second night at the Gambier Lake campground and hiked back across to West Bay the next day. This is where Sylvia took the additional samples from McDonald Creek, as the trail crosses into the head of West Bay.

A:

B:



Figure 5: Sam and Sylvia hiking up Gambier Creek with our camping and sampling gear (A). Sample site along Gambier Creek (B).

Long Creek

Long Creek also required boat access, so Kevin Wing dropped us off at the mouth of the creek, which comes out into Long Bay. The field team was Sam, Sylvia, and Sk̓wx̓wú7mesh Úxwumixw/Squamish Nation ACE Monitor, Jonny Williams. The community at the co-op was very welcoming, especially considering that we essentially docked the boat in the front yard of one family. They pointed us in the right direction and gave some helpful tips on which trails to follow. We sampled at three sites, following the trail to Lost Lake. Unfortunately, the creek dried up near the top here as well. In general, there was very little water flow, thus it was challenging to find good sample sites.

It was on this day that our pump broke, thus we were unable to take our fourth sample as the pump stopped working out of the blue. We took the pump apart and re-wired a couple of faulty connections. Luckily, we were able to fix it ourselves without having to send it back to UNBC for help.

A:



B:



Figure 6: Sam taking measurements of the residual depth pool (A). ACE Monitor Jonny Williams (B).

Center Creek

Sampling at Center Creek proved to be our most challenging day, due to unexpected issues with private property. The head of Center Bay is all private property, and the creek mouth was closed off with a log boom and is inaccessible to any boats. The only way we could access the creek was to moor our boat on a private dock. We met with the landowner and after explaining the project intentions, were granted access to sample at three sites and were accompanied by the landowner. Unfortunately, the sites were relatively close together. Center Creek was beautiful, flowing wonderfully and has sections of cascades, appeared to provide ideal CTF habitat.

Additional sampling in Grennon Creek, New Brighton Creek, and Mannion Creek

As we had 17 sample kits remaining at the end of summer, we decided to continue sampling during Fall and Winter. After including blanks and duplicates in our protocol we collected eDNA from nine more sites. These included six sites during October; four sites in Grennon Creek, and one more in Mannion Creek, and one in New Brighton Creek. Sampling during December included three sites located in two Whispering Creek tributaries. All sites were distributed locally within the southwest peninsula, among fairly dense residential developments. Grennon Creek and New Brighton Creek have developments throughout most of their riparian areas, including a main road along most of its length and many houses built within the riparian areas. In Grennon Creek, we sampled three sites within the relatively disturbed lower reaches and one site upstream where old forest riparian remains relatively undisturbed. Similarly, we sampled one site near the mouth in New Brighton Creek, which drains through extensive domestic developments and often runs dry in many places along its length during the summer. We expected the human disturbances in these streams might have displaced the CTF and therefore, our eDNA results were expected to fail to detect presence at all five sites. Similarly, sampling in two

Whispering Creek tributaries that enter downstream of a waterfall have extensive land developments relative to upstream and might preclude the presence of tailed frog.

Preliminary eDNA results and brief discussion.

The final eDNA results came in from the laboratory (Brent Murray Lab, UNBC) mid-March and are currently at the early stages of analyses. These initial interpretations are presented here to provide context for this field summary report. A complete analysis will be presented in a separate final report, the development of which is under way.

Briefly, the eDNA results from 52 sample sites distributed among nine streams detected the presence of CTF eDNA in 64% of sites, and these positive detections were clustered within five streams (Table 1; Figure 7). Negative results were clustered within four streams. Relatively dense human developments in New Brighton Creek, Grennon Creek, and Long Bay Creek suggest habitat disturbances can explain the absence of CTF eDNA. Similarly, the trends of eDNA presence and absence in Whispering Creek and Andy’s Bay Creek appear to be associated with human disturbances and loss of riparian areas resulting from domestic developments and industrial logging activities. Absence of CTF eDNA in samples from Center Creek might suggest something other than human disturbances have influenced the distribution of CTF on Gambier Island. Further analyses linking the CTF distribution with their biology and habitat use, and possibly including metrics of disturbance associated with the streams, will help address hypotheses that inform a conservation (Risk) assessment of this unique Gambier Island CTF metapopulation.

Table 1: Distribution of eDNA sample sites in Gambier Island streams collected during Summer 2021, and Summer, Fall, and Winter 2022. Duplicate results in Whispering, Mannion, New Brighton, and Grennon creeks are not shown, but see text.

Watershed	Total eDNA Sites	CTF eDNA Detected
MacDonald Creek	9	8
Whispering Creek	17	12
Mannion Creek	8	7
Andy's Bay Creek	3	3
Gambier Creek	4	3
Long Bay Creek	3	0
Center Creek	3	0
Grennon Creek	4	0
New Brighton Creek	1	0
Total Samples	52	33

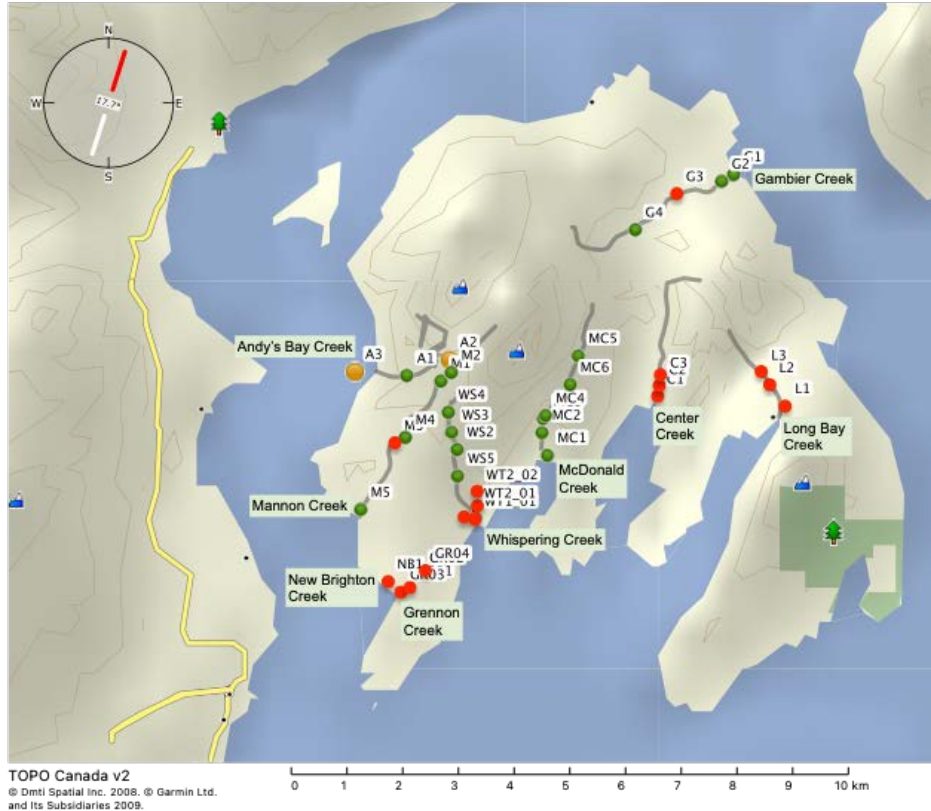


Figure 7: Map of Gambier Island showing the distribution of eDNA sample sites where Coastal Tailed Frog eDNA was detected (GREEN Dots), NOT detected (RED Dots) during Summer, Fall, and Winter of 2022. Two ORANGE Dots in Andy's Bay Creek highlight 'Low-Possible' detection.

Summary

The eDNA sampling has provided distribution data to begin addressing hypotheses concerning: 1) timing of colonization, 2) interactions with other species, and 3) human caused habitat disturbances. More generally, the eDNA results inform conservation management objectives by directing future actions toward the five streams where the species has been detected on Gambier Island. The brief description of results presented above support the hypotheses: 1) a wide distribution among streams supports the hypothesis that CTF colonized Gambier Island long ago and have survived in isolation for thousands of years since glaciers receded; 2) Multiple positive eDNA results within fish bearing streams suggest that CTF have adapted to sharing habitats with native fish populations in at least three streams; 3) human disturbances appear have caused range contractions in some streams and possibly explains absence in at least three streams. Limitations of eDNA data need to be addressed with follow up sampling to determine abundance estimates, a fundamental variable in determining conservation status. Furthermore, genetic analyses are needed to inform demographic structure and evolutionary divergence, which are also key to evaluating extinction risk to biodiversity.

One unexpected challenge encountered in the sampling process was insufficient water flows in some of the sampling sites. This made taking samples challenging in certain areas, and impossible in others. On a couple of occasions, we collected fewer samples than the originally planned after flowing the creek upstream to find it going subsurface near or before the headwaters. The sample sites in the fall were all found to have sufficient water flow, suggesting that at other times in the year there is ample water flow for eDNA sampling. These field observations, together with the habitat variables collected in the field will be incorporation with eDNA results and interpretations, and discussed in the next final report.

Of the nine sampled creeks, five have suitable CTF habitats and four appear to not support CTF. Among those streams where CTF were not detected, three have undergone human disturbances, such as the development of roads and houses, or are generally high traffic areas. The fourth negative eDNA stream (Center Creek) remains relatively undisturbed, however, which suggests something other than human disturbances also limit the presence of CTF in watersheds. The results overall suggest, however, that the distribution of CTF on Gambier Island has been affected by human activities, which include industrial logging, and domestic developments in their home streams. Consequently, conservation actions are needed to improve protection of streams where they still occur if we hope to avoid extirpations in the future.

A recommended next step toward informing the conservation assessment of this unique CTF metapopulation is to continue with follow up abundance sampling in streams where eDNA has detected their presence. Field methods have already been developed by BC Government biologists that inform abundance estimates and monitoring for CTF around human disturbances (Malt et al. 2014) and these same methods can be implemented here to determine the relative abundance of CTF among streams. The sampling includes capture and handling of frogs, and collections of habitat variables, and will therefore identify life histories present, further address hypotheses concerning habitat impacts from human disturbances, interactions with fish among streams. The methods require the strategic capture and handling of frogs and therefore facilitates the collection of non-lethal tissue samples for genetic analyses. Genetic data are needed to inform hypotheses about the timing when CTF colonized Gambier Island, demographic structure among streams, which also informs the risks and threats, including those associated with climate change.

Acknowledgments

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