# **Environmental DNA Field Sampling Summary-2022**

# Progress report for: Gambier Island Coastal Tailed Frog Distribution Project

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For

# Howe Sound Biosphere Region Initiative, Islands Trust Conservancy, and Gambier Island Conservancy.

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## Introduction

The Coastal Tailed Frog (Ascaphus truei) sampling 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 this 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, so that professionals working in the resource sector are aware of the distributions of SARA-listed species and to 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'ka7tsem/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 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) 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 Grennon Creek, Mannion Creek, Andy's Bay Creek, and New Brighton Creek where there was some degree of human disturbance and which could potentially no longer support CTF and therefore, would show absence in our eDNA samples. 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**

Our initial proposal planned for the sampling from seven of the largest watersheds on Gambier Island. We budgeted for a total of five sites per creek to evaluate influences from tributaries on eDNA distribution within each watershed. Planned sample sites included one located near the mouth of each creek, and then additional sites distributed upstream and spaced roughly 1.5km apart to evaluate the distribution of habitat use within each stream. Blanks consisting of distilled water filtered through the apparatus 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 detected at sites when they are actually absent). For field planning, we plotted proposed sample locations within the creeks on maps and considered the length of the creek and any known major tributaries. However, final sample site selection occurred in the field at locations where sufficient flows and the presence of pools were thought likely to provide habitat for tailed frog tadpoles.

During our field days, we had to adjust and modify our sampling plan due to elements such as the inaccessibility of sampling sites, insufficient water flow to conduct eDNA sampling, and considerations of private property. For example, most of the major tributaries were either completely dried up (subsurface) or in standing pools. This often resulted in our collecting fewer sample sites at each creek than initially planned. Using our remaining sample kits, we decided to expand our sample range to include additional creeks, resulting in a total of nine creeks sampled.

We set up our field headquarters in West Bay on Gambier Island where we stored and maintained our equipment, charging supplies, and distilled water for the blanks. This was a convenient location to begin and end each day and prepare our field equipment and collect sampling blanks. The sampling team varied, as we were lucky to have often been joined by an ACE monitor from the Skwxwú7mesh Úxwumixw/Squamish Nation or 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 <u>Peter Scholefield</u>, <u>President of the Gambier Island</u>

<u>Conservancy</u>. 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.



Sylvia on our second McDonald Creek sampling day.

## Whispering Creek

The second sampling day was conducted by Sylvia and Sam. Whispering Creek is very significant to the community of West Bay as it is salmon-bearing, is a 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 five samples from this creek to add to the previous sampling done in 2021.



Sam and our field set-up in Whispering Creek.

## Mannion Creek

The field team for Mannion Creek included Sam, Sylvia, Mike, and a Skwxwú7mesh 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 a very long creek with difficult terrain and no trails to follow, so we had to scramble up the creek. We took two samples, and at the second sample 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.



First CTF tadpole sighting.



Amy Baker, Mike Stamford, and Samantha Wing trying to access a difficult sample site in Mannion Creek.

A second field day was needed for Mannion Creek due to its terrain and size, so on our second day, we drove in through the logging roads, and then hiked to a different location on the creek. There we took two additional samples, resulting in a total of four sample sites at Mannion Creek.

### Andy's Bay Creek

Sam, Sylvia, and Mike sampled 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 several hours. We did successfully get to the sample site, which was 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 do the last samples at Mannion and Andy's Bay mouths 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.



Top: Sam and Sylvia hiking up Gambier Creek with our camping and sampling gear. Bottom: Sample site along Gambier Creek.

### Long Creek

Long Creek is also boat access only, 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 Skwxwú7mesh 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.



Top: Sam taking measurements of the residual depth pool. Bottom: ACE Monitor Jonny Williams

### 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, it seemed to have ideal CTF habitat.

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

As we had some sample kits remaining, we decided to sample at six additional sites including four in Grennon Creek, one more in Mannion Creek, and one in New Brighton Creek. Grennon Creek is near New Brighton and is heavily disturbed by human developments, including the main road along most of its length and many houses built within the riparian areas. We sampled from three sites within the disturbed lower reaches and one site upstream in a relatively undisturbed old forest riparian. Similarly, we sampled one site near the mouth of the adjacent New Brighton Creek, which drains through extensive domestic developments and often runs dry in many places along its length during the summer. We predict the human disturbances in these streams have displaced the CTF and our eDNA results will show absence at all five sites. We also added another site in Mannion Creek located downstream close to the mouth where sampling had not yet occurred.

Table 1: Number of eDNA samples collected from sites distributed among nine Gambier Island streams during Summer and Fall 2022. The number of samples collected does not include blanks but does include duplicates collected from one site each in Mannion Creek, Grennon Creek, and New Brighton Creek.

Creek	# Sample Sites	# Samples
McDonald	6	6
Whispering	5	5
Mannion	5	6
Andy's Bay	3	3
Gambier	4	4
Long	3	3
Center	3	3
Grennon	4	5
New Brighton	1	2
	Total: 34	Total: 37



Figure 1: Map of Gambier Island showing the distribution of eDNA sample sites (Red Dots)

#### collected from nine streams during Summer and Fall 2022.

#### Summary

The 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 from this sampling are intended to inform future conservation management objectives with presence absence data for Coastal Tailed Frog within and among streams of Gambier Island.

One unexpected challenge encountered in the sampling process was insufficient water flow in some of the sampling sites. This made taking samples challenging in certain areas, and impossible in others. On a couple of occasions, we had to take less than the originally planned amount of samples, as when we followed the creep upstream some of them dried up near or before the headwaters. In the future, sampling should be conducted earlier or later in the summer to avoid the driest season. 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.

Of the nine sampled creeks, seven appeared to have suitable CTF habitats at various points along the creek. The two which did not (Grennion and New Brighton) have undergone extreme human disturbances, such as the development of roads and houses, or are generally high traffic areas. The results of this research will be informative of the resilience of the CTF to human activities, such as through comparing the presence and absence of CTF in high and low disturbance areas. This type of information will be a key asset in the development of conservation management plans of these watersheds.

Possible future research includes sampling additional creeks for CTF or the expansion of the project to investigate additional at-risk species on Gambier Island, in creeks, wetlands, and woody areas. Continuation of this project will expand the knowledge of biodiversity on the Gambier Island allowing for a more comprehensive understanding of what we need to protect and what activities have negative outcomes for the survival of these species.

#### Acknowledgments

We acknowledge that Chá7elkwnech/Gambier Island is within the traditional territory of Skwxwú7mesh Úxwumixw/Squamish Nation and are grateful for their support from Rights and Title Department. Rachel Munger facilitated field assistance and monitoring for ACE Monitor's Amy Baker, and Jonny Williams, who also shared valuable insights into Skwxwú7mesh Úxwumixw perspective of the land and connections with Chá7elkwnech. Peter Scholefield (President, Gambier Island Conservancy) accompanied in the field to observe sampling techniques and shared GIC sampling equipment. More generally, Gambier Island Conservancy directors were united with Gambier Guardians in the opposition of logging old forest in Whispering Creek watershed and endorsed the use of CTF eDNA sampling to gather data necessary to evaluate impacts of logging on biological diversity. In 2021, Gambier Island Conservancy also provided funds to enable independent environmental and archeological assessments in Whispering Creek headwaters by Micaele Florendo (Maddison Consultants Ltd.) and Ecologic Environmental Consultants Ltd. who both kindly provided their professional services at cost. From the initial creation of this project, Mike Stamford has provided countless hours of time in support of this research. From connecting Sylvia and Samantha with various experts and contacts, guiding challenging field days and teaching us about eDNA and sampling protocol, to the writing of various proposals and reports, Mike has provided a great deal of in-kind services and time and has been essential in the creation and execution of this project.

Funding for this research was initiated in 2021 when local community members concerned with socioeconomic impacts on biodiversity on Gambier Island donated funds to the Gambier Island Conservancy (GIC), which were used for preliminary assessments of CTF in Whispering Creek. Subsequently in 2022, Howe Sound Biosphere Region Initiative Society (HSBRIS), Islands Trust Conservancy (ITC), and Habitat Conservation Trust Foundation (HCTF) provided funds to expand scope.

# References

B.C. Conservation Data Centre. 2016. Conservation Status Report: Ascaphus truei. B.C. Minist. Of Environment. Available: <u>https://a</u>100.gov.bc.ca/pub/eswp/ (accessed Dec 1, 2020).
Carim, K., McKelvey, K., Young, M., Wilcox, T., Schwartz, M. 2016. A protocol for collecting environmental DNA samples from streams. Gen. Tech. Rep. RMRS-GTR-355. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 18 p.

- Ecologic Consultants Ltd. 2021. Whispering Creek watershed assessment. *For* Gambier Island Conservancy.
- Environment and Climate Change Canada. 2016. Management Plan for the Coastal Tailed Frog (Ascaphus truei) in Canada [Proposed]. Species at Risk Act Management Plan Series. Environment and Climate Change Canada, Ottawa.2 parts, 3 pp. + 49 pp.
- Forest Practices Board. 2014. A decade in review: Observations on regulation forest and range practices in BC. Special Report FPB/SR/46.
- Gambier Forest Products Ltd. 2020. Woo39-Harvest Instructions and site plan for KI-104. Overland Resources Ltd. October 2020.
- Hobbs, J., Round, J. M., Allison, M. J., & Helbing, C. C. (2019).
  Expansion of the known distribution of the coastal tailed frog, Ascaphus truei, in British Columbia, Canada, using robust eDNA detection methods. PLOS ONE, 14(3), e0213849.
  <u>https://doi.org/10.1371/journal.pone.0213849</u>
- Hayes, M.P., T. Quinn, D. J. Dugger, T. L. Hicks, M. A. Melchiors, and D. E. Runde. 2006. Dispersion of coastal tailed frog (Ascaphus truei): a hypothesis relating occurrence of frogs in non-fish-bearing headwater basins to their seasonal movements. Journal of Herpetology 40(4):533-545.
- Maddison Consultants Ltd. 2021. Preliminary sensitive species assessment for cutblock 39 at the headwaters of Whispering Creek, Gambier Island, British Columbia (BC). www.maddisonconsultants.com
- Madrone Environmental Consultants, LTD. 2009. Terrestrial Ecosystem Mapping (TEM) for Howe Sound Islands – Bowen, Gambier, Keats, Anvil and Other Associated Islands. Islands Trust Fund.
- Stamford MD, Strohm J., Euchner T, Pitt C, and B Murray. 2022. Williston Grayling distribution: Parsnip, Peace, Dinosaur. FWCP Project Number: PEA-F21-F-3198.
- Wahbe TR, Ritland C, Bunnell FL, and K Ritland. 2005. Population genetic structure of the tailed frogs (*Ascaphus truei*) in clearcut and old groweth stream habitats in south coastal BC. Can J. Zool. 83:1460-1468.