BC Ministry of Forests, Lands and Natural Resource Operations Coast Area Research Section 2014/15 Wildlife Research Project Summary

The Spatial Ecology of Coastal Tailed Frogs in Northwestern BC



Photo: A. McEwan

Kalum Tailed Frog Research Team

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Background

Coastal Tailed Frogs (*Ascaphus truei*) are blue-listed provincially (species of special concern); an Identified Wildlife Species under the Forest and Range Practices Act (FRPA); a focal species for Ecosystem-based Management (EBM) in the Central and North Coast, and South Central Coast, Land Use Orders; and a Schedule 1 species of Special Concern under the federal Species at Risk Act. Though tailed frogs are widely distributed throughout their range, their unique taxonomic nature and metamorphic life history, in combination with their reliance on both aquatic and terrestrial headwater stream habitats vulnerable to the cumulative effects of land use, result in the current conservation status of this species in BC.

The Ministry of Environment B.C. Conservation Framework identifies Coastal Tailed Frogs as a priority 1 species under the goal of "preventing species and ecosystems from becoming at risk" (Goal 2).

- Tailed frogs have the longest lifespan (15-20 years), larval period (1-4 years), and age to sexual maturity (7-8 years) of all North American frogs; they depend on cool, fast-flowing mountain streams with stable, step-pool morphologies, and they depend on them for a very long time. Where they occur, they are excellent indicators of headwater stream integrity.
- Coastal Tailed Frogs are widely distributed and commonly found through the geographic range of the species, but they are not present in all watersheds. Climate, bedrock geology and physiography (e.g., basin size and steepness) determine where populations will be found. Where they are found, population distribution and abundance is determined by forest and stream conditions sensitive to land use practices (e.g., sedimentation, changes in stream temperature; modified riparian vegetation or upland forest).
- Juvenile and adult frogs have narrow temperature tolerances (~6-18 °C) and are highly susceptible to desiccation. In the central and northern portions of their range in BC, they appear to spend a substantial part of their adult life away from streams in cool, moist micro-sites and habitats closely associated with old growth and mature second growth forest.

To date, most research to support tailed frog conservation guidance has concentrated on aquatic larval (tadpole) populations, studying the effects of land use on flow, water quality, and stream channel and bank integrity (e.g., Dupuis and Steventon 1999). This limits our understanding of the sensitivity of tailed frog populations to land use, since frogs spend only a quarter, or less, of their lives as tadpoles. There is evidence that the densities and productivity of aquatic larval stages are limited by different factors than those affecting juveniles and adults living in adjacent terrestrial areas (Mallory and Richardson 2005; Matsuda and Richardson 2005). Reductions in terrestrial habitat condition and concurrent reductions in adult and juvenile survival might go undetected adjacent to what appear to be productive larval streams. Conservation measures should capture important terrestrial habitats and features, as well as aquatic in-stream values.

Cumulative effects of land use can potentially degrade riparian and upland forest habitats beside streams, and disrupt movement patterns and isolate tailed frog populations due to movement barriers (e.g. roads and/ or large stretches of young forest). It is necessary to understand the scale at which conservation management will be most effective. The distribution of conservation management efforts across landscapes requires an understanding of the connectivity of, and genetic exchange between, local populations of tailed frogs, mediated by the dispersal and movements of juvenile and adult frogs through aquatic and terrestrial habitats within and between stream basins.

There are several knowledge gaps in our understanding of the ecology and life history of tailed frogs; in particular, for the juvenile and adult phases. A 2009 review of tailed frog research and inventory needs for the Ecosystem-based Management Land Use Orders for the Central and North, and South Central Coast, identified priorities such as radio-telemetry and the study of genetic connectivity within and between populations, as well the need to determine the optimal spatial configuration for tailed frogs may be integrated at a range of scales with management for riparian, biodiversity and wildlife habitat values in headwater stream systems, at the top of the stream continuum, benefiting downstream resources (e.g., high value fish streams).

Study Goal

Describe and quantify the spatial ecology of the Coastal Tailed Frog in northwestern BC, determining patterns of habitat use, movement and dispersal, population density, and population distribution at multiple relevant scales for all life history phases of this metamorphic amphibian.

Projects and Timelines

There are a number of related projects underway, each with their own objectives, designed to address the overall study goal. The study began with a FLNRO/MOE feasibility pilot and site establishment project in 2010. Projects have various funding and partner agencies, project leads, and project durations (see Table 1). A glossary of agency acronyms is included in Appendix A.

1. The Habitat Ecology of Coastal Tailed Frogs: terrestrial habitat requirements of postmetamorphic Frogs (2011-2014). FLNRO/MOE/UNBC/HCTF.

Objective: quantify patterns of terrestrial habitat use and movement for frogs using radiotelemetry (a first for this species) and more traditional survey methods.

2. Factors influencing the abundance and distribution of Coastal Tailed Frogs (2011-2014). *FLNRO/MOE/UNBC*.

Objective: quantify relationships between tailed frog populations, habitat and microclimate, within the stream to streamside to upland gradient.

3. Conservation effectiveness evaluation (2011-2014). FLNRO/MOE

Objective: develop and test aquatic and terrestrial field protocols and indicators to monitor the effectiveness of tailed frog conservation measures, such as Wildlife Habitat Areas and riparian Best Management Practices.

4. Efficacy of survey techniques for the detection of juvenile and adult Coastal Tailed Frogs (2012-2015). FLNRO/UNBC

Objective: assess the sensitivity of post-metamorphic survey techniques (pitfall activity traps, visual encounter surveys, passive cover boards (used by the Northwest Regional Amphibian Monitoring Program (2011-12 HCTF Project; Kerby et al.)) and assess their practical utility in inventory and monitoring for tailed frogs.

5. Reproductive ecology of Coastal Tailed Frogs (2013-2015)

Objective: Assess the role of key seasonal reproductive habitats (i.e., locations for breeding aggregations, egg-laying and over-wintering) in tailed frog populations.

6. Detection and prevalence of the Chytrid fungus (*Batrachochytrium dendrobatidis*) (2011-2013). MOE/FLNRO

Objective: document the presence and distribution of *Batrachochytrium dendrobatidis*, the causal agent of the amphibian disease chytridiomycosis, in tailed frog and western toad (*Anaxyrus boreus*) populations (both Species-at-Risk) of northwestern BC.

7. Fringe science: determining the range extent of tailed frogs in northwestern BC (2011-2017). *FLNRO/MOE/UNBC*

Objective: identify and assess fixed and variable factors influencing the distribution of tailed frogs at the northern edge of the range of the species.

8. Molecular detection of stream-dwelling amphibians in headwater streams (2012-2014). *FLNRO/UNBC*

Objective: explore the efficacy and utility of using environmental DNA (eDNA) from water samples to provide a rapid assessment of tailed frog occupancy and distribution within streams, watersheds and landscapes.

9. Genetic connectivity across Coastal Tailed Frog landscapes (2014-2017). UNBC/FLNRO

Objective: assess landscape genetics and population connectivity to support conservation strategies which can address the cumulative effects of land use and climate change at multiple scales.

Project	2010	2011	2012	2013	2014	2015	2016	2017
Pilot: Methodologies and Site Selection	Y							
1. Habitat Ecology - Terrestrial		Y	Y	Y	Y			
2. Factors - Abundance and Distribution		Y	Y	Y	Y			
3. Conservation Effectiveness Evaluation		Y	Y	Y	Y			
4. Survey Techniques			Y	Y	Y			
5. Reproductive Ecology				Y	Y			
6. Chytrid Distribution and Prevalence		Y	Y	Y	Y			
7. Northern Range of Tailed Frogs		Y	Y	Y	Y			
8. Environmental DNA (eDNA)			Y	Y	Y			
9. Landscape Connectivity					Y	Y	Y	Y

Table 1. Project field schedule

Established research sites provide opportunities for long-term population and monitoring studies (e.g., climate change, cumulative effects). Pitfall traps will be capped and secured at the end of the study, and kept in place for monitoring and future research.

Study Location and Design

A substantial body of tailed frog research, inventory and assessment work had been completed in the Coast Mountain Resource District (Kalum and North Coast Timber Supply Areas) of northwestern BC. Kalum Coastal Tailed Frog research and inventory undertaken from 1994 to 2002 (multiple references, e.g. Dupuis & Steventon 1999; Dupuis & Friele 2003) provided the basis for site selection in this current research study, as well as a unique opportunity to compare tailed frog habitats and populations over a 10-20 year period in managed landscapes.

Details of the current design (Todd et al. 2010) include:

• 15 randomly selected research sites drawn from a pool of known occupied sites (based on the 1994-2002 inventory and research) blocked by treatment type (3) and watershed (5) (Figure 1);



Figure 1. Study area: 15 sites (white/black circles) distributed across five Skeena River watershed blocks located northeast of Terrace, BC. The red line indicates the previous northeast range boundary for the species (Dupuis and Friele 2003), and is currently under revision.

- 3 treatment types within cut-blocks from 5 to 20 years old: 1 undisturbed (no harvesting within 100m of stream); 2 50m retention buffer (clear cut harvest beyond buffer); and 3 harvested (clear-cut to stream);
- 4 of the 5 study watersheds contain Coastal Tailed Frog Wildlife Habitat Areas (WHA): Ascaphus (AS), Gosling (GO), Kleanza (KL), and Zymoetz (ZY);
- the 5th study watershed Copper (CS) is in the headwaters of the Zymoetz River, anchored on streams found to be occupied by Tailed Frog adults within the Bulkley TSA (Steventon 2009), beyond the presumed range boundary of the species proposed by Dupuis and Friele (2003) (Figure 1);

- frog radio-telemetry and pitfall trapping take place in the GO, AS and KL sites, and
- visual encounter surveys for frogs are integrated with habitat surveys, microclimate monitoring, and tadpole and stream monitoring at all sites in all 5 watersheds.

Field Activities

Activities are guided by established field method protocols revised annually (Todd et al. 2014) and/or specific to graduate research (e.g., McEwan, In Prep) and include, but are not limited to, the following:

Frogs and Tadpoles

At intensively studied research sites in the Gosling (GO), Ascaphus (AS) and Kleanza (KL) watersheds (2011 to 2014):

- frogs are located using visual encounter surveys (VES), pitfall traps and cover boards to provide estimates of relative density and distribution;
- captured frogs are measured and marked (visual implant elastomer) for a long-term population mark and recapture study;
- frogs of sufficient size are fitted with a radio transmitter/belly belt attachment and relocated daily (the first use of radio-telemetry with this species)
- for the study of habitat use, up to 20 frogs/year were followed for between one and two weeks (maximum transmitter life of 14 days); each relocation is considered a 'used' site and a paired 'random' site is located within 3-20m of the used location to determine if frogs are selecting for habitats or attributes;
- for the study of movement patterns, up to 10 frogs/year are recaptured and refitted with new transmitters every 2 weeks for up to 6 weeks;
- time-constrained stream searches (two observers for 15-minute intervals) are conducted annually (August low flow) in 100m-long stream reaches at all research sites to confirm continued tadpole occupancy, document tadpole population demographics (age cohort, length, weight), and provide relative estimates of tadpole abundance adjacent to adult terrestrial habitats.

At the sites on tributaries to the mid-Zymoetz River (ZY) (2011 to 2013) and upper (CS) Zymoetz River (2011-2012):

- visual encounter surveys (VES) are used to provide estimates of frog density and distribution
- captured frogs are measured and marked (visual implant elastomer) for a long-term population mark and recapture study;
- time-constrained stream searches (two observers for 15-minute intervals) are conducted annually (August low flow) in 100m-long stream reaches at all research sites to confirm continued tadpole occupancy, document tadpole population demographics (age cohort, length, weight), and provide relative estimates of tadpole abundance adjacent to adult terrestrial habitats.

Microclimate and Habitat

Stand-level terrestrial microclimate and habitat monitoring arrays (8 stations per array; iButton data loggers) are installed at all 15 sites, recording soil, surface and air temperature and relative humidity throughout the growing season. HOBO data loggers record air temperature throughout

the winter. Vegetation and structural habitat plots are completed at each microclimate station and pitfall trap array.

HOBO TidBit data loggers are installed at all sites to monitor stream temperature continuously through the year. Aquatic habitat condition, stream geometry and morphology are characterized annually at low flow in 100m stream reaches adjacent to the terrestrial study sites.

Fringe Inventory

Time constrained larval surveys (TCS), following the study's standard 2-observer 15-minute (total of 30 minutes) protocol, were completed for the tributaries of the upper Zymoetz River within the Bulkley TSA in 2011. The objective was to map out the extent of the species range beyond the previously recognized range boundary (see Figure 1), and provide some input into BCTS operations in this area.

Low detection rates raised concerns about the possible ineffectiveness of reconnaissance level TCS for the detection of larval occupancy in areas of low larval density. This prompted the exploration of more intensive survey methods in Year 1 (e.g., area-constrained searches), concluding that the level of survey effort required to adequately map frog distributions in these low density streams was extremely costly and beyond the scope of this project. Less intensive, more cost effective methods, such as environmental DNA (see above) are under investigation.

<u>Chytrid</u>

Up to 30 juvenile and adult frogs and 30 toads captured in each watershed are swabbed each year as per the MOE (2008) protocol

(http://www.env.gov.bc.ca/wld/documents/wldhealth/Collection%20Protocol.pdf). Swab samples are analysed by the Animal Health Laboratory (Abbotsford) to detect the presence of *Batrachochytrium dendrobatidis*, also called the chytrid the fungus.

Reproductive Ecology

In 2011, autumn breeding aggregations were discovered in two of the study sites (the first such observation in BC). These locations are monitored in fall and spring - visually and using remote cameras - to determine the number of frogs using sites, the timing of breeding, and the potential use of these sites for over-wintering. Fall and spring radio-telemetry and frog surveys (pitfall traps and VES) are also used to determine movement to and from key reproductive locations.

Genetics and eDNA

Larval voucher specimens were collected from all study sites in 2011 to provide genetic markers.

Markers are used to identify environmental DNA fragments in water samples to detect the presence of tailed frog in streams (Goldberg et al. 2011). 2012 water samples were taken, using 2 sampling methods at all 15 research sites and eDNA presence and concentrations were quantified using qPCR. Sampling methods were compared, and eDNA concentrations were compared to larval densities from time-constrained searches. 2013 sampling was expanded to an additional 30 random sites to further quantify the relationship between eDNA concentrations and tadpole densities, and 2014 will examine the influence of stream flow and other factors on eDNA concentrations.

Beginning in 2014, a UNBC PhD student will begin more broadly distributed genetic sampling of tadpoles (small tail clips) and frogs (skin and buccal swabs) to explore the connectivity of populations between watersheds and across landscapes.

Project Deliverables

Most projects are in their last field year for 2014 (Table 1). Annual progress reports have been submitted to the Habitat Conservation Trust Foundation (Todd and Johnson, 2012, 2013 and 2014). A brief summary of progress is presented in Appendix B. A summary of extension to date is presented in Appendix C.

Final products will include graduate and undergraduate theses, peer-reviewed scientific technical reports, published manuscripts and conference presentations, as well as non-technical extension products including workshops and extension notes. Research results will provide recommendations for improved guidance for the management and monitoring of tailed frog populations and habitats, and riparian and upland forests associated with headwater streams. Such guidance will be useful to the assessment, management and monitoring of Coastal Tailed Frogs and headwater streams undertaken through regulation, legislation and processes such as: BC Forest and Range Practices Act and Forest Planning and Practices Regulation; Forest Resource Evaluation Program (FREP); Riparian Areas Regulation; Species at Risk Act and BC Wildlife Act; BC Environmental Assessment Act; and provincial Best Management Practices (e.g., forestry, run-of-river independent power projects, linear developments, urban development). Research will improve predictive modeling of tailed frog distributions in coastal BC, and the cumulative assessment of land use impacts and changing climate to tailed frogs and headwater streams.

Resources and Manpower

The Kalum Coastal Tailed Frog Study is a joint-region, joint-agency research effort initiated by the Research Sections of the Coast Area and Skeena Region, and the Skeena Region Ecosystems Section, of the BC Ministry of Forests, Lands and Natural Resource Operations, and the Ministry of Environment. The Study is co-led by the University of Northern British Columbia.

Study Leads:

Melissa Todd (MFLNRO Coast Area Research; Research Wildlife Ecologist) and Dr. Chris Johnson (UNBC Ecosystem Science and Management; Associate Professor; spatial and population ecology).

Research Team includes:

- Dr. Purnima Govindarajulu (MOE Conservation Science Section; Amphibian/Reptile/Small Mammal Specialist)
- Kathy Paige (MOE Ecosystem Conservation; Ecosystems Biologist and FREP Wildlife Resource Value Team co-lead)
- Anne Hetherington (MFLNRO Ecosystems Section Skeena Region; Ecosystem Biologist)
- Len Vanderstar (MFLNRO Ecosystems Section Skeena Region; Ecosystem Biologist)
- Laurence Turney (Ardea Biological Consulting; Wildlife Biologist)
- Lis Rach (TerraNiche Environmental Solutions; Wildlife Habitat Ecologist)
- Dr. Brent Murray (UNBC Ecosystem Science and Management; Associate Professor; molecular ecology)
- Doug Steventon (Retired FLNRO Skeena Research Wildlife Ecologist)

Graduate Students

Alexis McEwan, UNBC MSc candidate supervised by Dr. Chris Johnson; M. Todd (FLNRO) and Dr. P. Govindarajulu (MOE) are committee members; fine scale habitat selection / radio-telemetry; in the field from 2011 to 2012, and with summer assistants, supported by HCTF grant and FLNRO project grant.

Cherie Mosher, UNBC PhD candidate co-supervised by Dr. Chris Johnson and Dr. Brent Murray; M. Todd (FLNRO) is committee member; population genetics study; field work starting in 2014; supported by NSERC Discovery Grant (Johnson); the UNBC Biodiversity Monitoring and Assessment Program (Johnson and Murray); and FLNRO project grant.

Funding

The Kalum Coastal Tailed Frog project is primarily funded by:

- BC FLNRO Natural Resource Sector Research Project funds (Todd);
- Habitat Conservation Trust Foundation, Habitat Enhancement Program Grant (Todd and Johnson)
- MOE Ecosystem Conservation and Conservation Science program funds (Paige and Govindarajulu);
- NSERC Discovery Grant (Johnson);
- UNBC Biodiversity Monitoring and Assessment Program (Johnson and Murray).

<u>Project Partners</u> provide substantial in-kind support in the form of field safety check-ins, logistical and equipment support, field volunteers, and site access:

- BC Timber Sales Skeena Business Area
- BC MFLNRO Coast Mountains Resource District

Site access and support is also provided by:

- Coast Tsimshian Resources / Brinkman Forest Ltd.
- Northwest Timberlands (on behalf of Kitselas Forest Products)
- BC Timber Sales Babine Business Area

<u>Public Volunteers</u> have donated critical support to the field effort, including Norma Kerby, Amanita Coosemans, Ken Adair, Grant Hazelwood, Gina LaHaye, Irene Vanderstar, Hamish MacMillan, and Jeff Meggs.

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Appendix A. Agency Acronyms

FLNRO	BC Ministry of Forest, Lands and Natural Resource Operations
HCTF	Habitat Conservation Trust Foundation
MOE	BC Ministry of Environment
UNBC	University of Northern British Columbia

Appendix B. 2010-2013 Progress

The following is a brief summary of progress and preliminary results based on available data and limited analyses completed to date, as reported in Todd and Johnson (2012, 2013, and 2014). Interim progress is descriptive; results of statistical analyses and modeling will be presented along with interpretations and conclusions final reports and publications currently in prep. Preliminary results presented below may be amended based on analyses of final 2011-2014 datasets.

- 1. The Habitat Ecology of Coastal Tailed Frogs: terrestrial habitat requirements of postmetamorphic Frogs (2011-2014). FLNRO/UNBC/MOE/HCTF.
- **2.** Factors influencing the abundance and distribution of Coastal Tailed Frogs (2011-2014). *FLNRO/MOE/UNBC*.
- 3. Conservation effectiveness evaluation (2011-2014). FLNRO/MOE

Data are collected jointly for these three projects, and analysed to address specific project objectives.

Analyses of 2011-2012 use-availability telemetry data (928 used and paired random locations from 38 frogs radio-tagged over 2 years), 2012 and 2013 pitfall trap results, and environmental and habitat variables are almost complete as part of MSc thesis preparation (McEwan, in prep).

- McEwan has devised a suitable statistical analysis, given the distribution of those pitfall capture data. She has attributed trap data to ecological (e.g., time of year, local climate, distance from stream, vegetation composition) and management variables (e.g., site treatment, canopy closure, distance from maternal stream) that may explain temporal and spatial variation in the capture of frogs. Those analyses are complete and a draft of the thesis chapter has been reviewed.
- The second set of analyses is focused on location data collected for adult frogs fitted with radio-transmitters during the first two field seasons. Frog locations and corresponding random sites have been attributed with ecological and management variables that may explain the movements and distribution of individual frogs. McEwan is using a species distribution model to identify key habitat attributes and features that are selected or avoided. Data preparation is complete and she is currently engaged in producing and evaluating candidate models that best describe the spatial ecology of radio-tagged frogs (as an index of the adult population).

In 2013, the research team placed 7 radio-transmitters (10 to 14-day lifespan) in the fall, tracking individual frogs daily for up to 5 weeks. Radio-tagged frogs were intercepted moving to their fall breeding locations; their point of origin prior to being trapped is unknown.

Preliminary results of species distribution modelling illustrate a significant affinity for large, decayed coarse woody material, root hollows, and moist micro-sites (e.g., devil's club draws, skunk cabbage seepage sites) located in riparian and upland forest (McEwan, in prep). Adult tailed frog populations appear highly mobile during critical seasonal periods dictated by their reproductive ecology (i.e., egg-laying, breeding), with the focus on movement to and from high quality reproductive stream and riparian forest habitats. In contrast to mid-summer, where daily movements were <15m, daily movements in early summer and autumn corresponding to periods of egg-laying and adult breeding, respectively, regularly exceeded 20m. The furthest distance moved between 2 days was 76m (gravid female in autumn). Results also indicate seasonal aggregations for early summer females laying eggs in high quality stream reaches, observed in

other studies elsewhere, and fall breeding aggregations, confirmed for the first time in this study. Pitfall trap and VES surveys at some sites produced relatively high juvenile frog capture rates in August and early September, thought to represent juvenile dispersal events.

Site-level frog distribution, as indicated from pitfall trap and VES surveys, is weighted towards streamside. Model evaluation in McEwan's (in prep) analyses of pitfall count data indicates that annual capture rates are sensitive to the interaction of trap distance from larval stream with time of year, as well as days since last rain (a proxy for ground moisture directly influencing frog activity). Survey results demonstrate a high level of activity in and around reproductive streams during early summer egg-laying and fall breeding seasons when frogs aggregate. However, these are periods of high mobility, during which adult frogs move relatively long distances to get to and from reproductive stream locations (up to 172m away from stream during monitoring).

In summer, when frogs appear to be less mobile, they are located up to 100m from the streams (survey areas only extend to 100m from the streams), but occur farthest from streams in undisturbed forest. Radio-tagged frogs in old growth and buffer sites were regularly caught and/or relocated between 30-100m from the stream. Frogs caught and radio-tagged >80m from the stream in summer were resident in these areas for the monitoring period, and were frequently relocated well beyond 100m from the stream. Conversely, those frogs radio-tagged in the recent clear cut sites were caught at and stayed close to the stream edge. The exception was the older regenerating Kleanza clear cut site (14.5km, Kleanza FSR; >20 years since harvest) with a high young tree density, producing a moister and more stable microclimate at ground level.

McEwan's (in prep) model evaluation using pitfall count data also suggests that variability in pitfall capture rates is related to forest age. Naive plots of tailed frog pitfall captures illustrate higher relative abundance in undisturbed forest (Figure 2). This is a similar trend to that reported by Hawkes and Gregory (2012), who found that Coastal Tailed Frogs in western Washington declined in clear-cut upland habitats 2- and 10-years following logging.



Figure 2. Pitfall trap captures by treatment type and gender for 2012 and 2013 combined (McEwan et al. 2013). Males and females are adults; unknown gender = juvenile and metamorph age classes.

In our current study, frogs located within clear-cuts during day-time visual encounter surveys in mid-summer were found under or within large woody debris.

Ambient air and ground surface temperature and RH, soil temperature, and stream temperature have been monitored at all sites to evaluate the effects of harvest treatments on frog habitat quantity and quality, and as potential explanatory variables for frog capture and telemetry data. Paired stations separated by 20m were positioned at 0m, 5m, 25m, 50m and 100m from the stream in 2010 and 2011 to evaluate differences between sites and years in stream temperature and microclimate conditions at varying distances from the stream. Preliminary examination of 2011 data indicated a shift in ambient air temperature and relative humidity somewhere between 5 and 25m, and again between 25 and 50 metres from the stream in the buffer sites, but not in the relatively homogenous old growth and clear cut sites. Stations were therefore added at 15m and 35m in the buffer sites in 2012 and 2013 to better assess the influence of edge and interior forest on a 50m buffer.

Structural and compositional habitat plots were completed for all microclimate stations. The sensitivity of microclimate variables, as measured using iButton dataloggers, to changes in habitat structure and composition is currently under analysis (UBC undergraduate thesis). Relationships will be modelled against harvest treatment type and frog abundance and distribution (entire dataset) to further quantify the influence of microclimate and habitat structure on tailed frog abundance and distribution, and evaluate the implications of retention buffers for tailed frog populations.

Since the start of the study, a total of 525 individual adult and juvenile frogs have been marked in the 9 most intensively studied sites (9 hectares in total) in the Gosling, Ascaphus and Kleanza watershed blocks (these watersheds have pitfall trap arrays installed and are part of the radiotelemetry study). Adult recapture rates in these 9 sites continue to be insufficient to support population estimates (2013 recaptures n=10; 6% of adult frogs marked in 2011 and 2012), though they are consistent with similar studies in southern BC (e.g., Wahbe et al. 2004, 6.7%; Matsuda & Richardson, 2000, <10%).

Annual time-constrained larval surveys were conducted for four years (2010-2013) at all 15 sites, along with continuous stream temperature monitoring and the annual measurement of a suite of aquatic habitat attributes. These surveys confirm continued reproductive occupancy of the research sites, and monitor larval population and stream habitat conditions at various treatment sites. High annual variability in larval relative abundance within sites results in high betweensite variability; sites with relatively high larval densities in one year can become low relative density sites the following year. Analyses are underway to evaluate factors influencing probability of larval detection and assess the effect of harvest treatments on aquatic component of the population.

Larval surveys have been performed primarily in mid-Summer (August) all years, and adult frogs are rarely detected. The first detection of an adult frog during a larval TCS was in 2012. Adults are detected in streams in early summer (females laying eggs) and fall (breeding season) during VES.

4. Efficacy of survey techniques for the detection of juvenile and adult Coastal Tailed Frogs (2012-2015). FLNRO/UNBC

Three methods have been used to detect adult and juvenile tailed frogs in the riparian and upland forests adjacent to reproductive streams: pitfall trap arrays, visual encounter surveys (VES) and cover-board arrays. Seasonal pitfall trap sessions (spring, summer, fall) were run in 2012 and

2013 at 9 sites. Systematic visual encounter surveys (VES) were completed each season at 15 sites in 2012 and 12 sites in the spring and fall in 2013. Cover-boards were in place by the start of 2013 at 6 sites only.

In 2012, pitfall traps caught more frogs than VES (126 versus 49, respectively), and was suggested to be the more effective method of survey. However, in 2013, pitfall capture rates in the Gosling and Kleanza were substantially lower (0.56 frogs /100 trap nights) than in 2012 (1.87 frogs /100 trap nights). Though this might suggest a change in population abundance, the reverse occurred for VES; capture rates increased in 2013 as compared to 2012. A total of 114 frogs were detected (71 in Gosling, 35 in Kleanza and 8 in Ascaphus sites) in 54 hours of survey in 2013, as opposed to only 49 frogs detected in 69 hours of survey in 2012. This suggests that population levels did not change between years, but the effectiveness of the two survey techniques had.

The timing of seasonal pitfall trap sessions differed by 2 to 3 weeks between years, affecting ambient and ground temperature and moisture. As previously mentioned, model evaluation in McEwan's (in prep) analyses of pitfall count data indicates that annual capture rates are sensitive to the interaction of trap distance from larval stream with time of year, as well as days since last rain (a proxy for ground moisture directly influencing frog activity). The latter 2 variables (time of year and days since last rain) are indicative of probability of detection in the survey since frogs are more mobile and vulnerable to trapping at different times of the year, and will be more active on humid days, again increasing their vulnerability to capture. These data illustrate the sensitivity of survey results to time of year, as a function of seasonal life history and climate (temperature and moisture), and daily differences in weather. Trap methodology in 2013 also changed slightly; a heavier gauge escape string for small mammal by-catch was used in pitfall traps and was assumed not to affect the chance of frog escape. Conversely, VES field crews were more trained in 2013 than in 2012 (first year of the survey) and, again, 2013 VES took place in different weeks within a season 'window' and under different daily weather conditions. A UNBC undergraduate thesis will model this inter-annual variability in VES and pitfall trap results against a suite of explanatory variables (e.g., date, temperature, humidity, trap method) to better understand the performance of each survey method.

Only 1 frog was captured beside a cover-board in 2013. Cover-board installations were completed in early 2013, and boards do need a substantial amount of time to settle and weather into their locations before they become potentially productive. A review of the current design of cover-board arrays relative to the objectives of their deployment is underway.

5. Reproductive ecology of Coastal Tailed Frogs (2013-2015)

In 2011 and 2012, autumn breeding aggregations were reported at two of the study sites (the first such observation in BC). Site monitoring and telemetry data suggested these are potential overwintering sites (Todd et al. in prep). In addition to gravid females and breeding males, nonbreeding frogs, juveniles and even metamorphs (just emerged from streams) arrived at breeding locations. Adult male and female frogs radio-tagged at these aggregations moved into and under large woody debris and did not emerge prior to snowfall. Breeding aggregations were located 32km apart on opposite sides of the Skeena River, and were located 30.6m and 12.8m from the main reproductive streams, situated on moist micro-sites anchored on ephemeral or sub-surface flow.

In 2013 we placed 2 Reconyx HC600 remote cameras at the larger of the 2 breeding sites in the Kleanza watershed to assess the utility of using remote cameras in determining the size of aggregations, the extent of the breeding period, and the use of these sites for over-wintering.

Photo review will be completed in 2014, but cursory inspection indicates that cameras have recorded high levels of frog activity at this location. Male frogs were observed engage in aggressive activity, suggesting potential competition for resources.

6. Detection and prevalence of the Chytrid fungus (<u>Batrachochytrium dendrobatidis</u>) (2011-2013). MOE/FLNRO

Over 3 years, all tailed frogs and western toads captured during the course of the study were swabbed (MOE 2008a) to determine if they were carrying the Chytrid fungus.

A total of 140 toad swabs were collected from 13 of the 15 sites (no toads were ever caught at sites KL-2 or CS-2) and analysed (DNA assay for presence of fungus; BC Animal Health Laboratory, Abbotsford, BC). 20 swabs were positive for Chytrid (14%). Positives were confirmed in 8 out of the 13 study sites, in 4 of the 5 watersheds (GO, AS, KL and ZY). The 5th watershed (CS) is comprised of low density sites located on the edge of the species' range and there was insufficient sampling completed to allow for any conclusions. None of the toads which swabbed positive for Chytrid showed any external signs of disease. This is consistent with a report by Deguise and Richardson (2009) in southwestern BC where 28% of 32 swabbed toads were positive for Chytrid, with no apparent affect on body condition. The presence of the Chytrid fungus on Western Toads north of 50 degrees latitude in BC is consistent with detections of Chytrid in Western Toads in the Northwest Territories (Shock et al., 2010)

Tailed frogs captured in 2012 and 2013 from all 15 study sites were swabbed for Chytrid; a subset of 203 swabs were analysed. All swabs came back negative for Chytrid. Sufficient swabs were collected in 4 of the 5 watersheds to conclude (~95% confidence; P. Govindarajulu, pers. comm.) that Chytrid is not currently present in the tailed frog population. Chytrid has been found on tailed frog adults in Oregon, where Hossack et al (2010) reported a low incidence of Chytrid in stream-dwelling amphibians in headwater streams. Three of 38 tailed frogs swabbed (8%) in that study were positive for Chytrid. Given the potential for tailed frogs to carry Chytrid, the unknown extent to which this fungus may cause disease in headwater stream amphibians at northern latitudes, and the presence of the fungus in toads caught in 4 out of 5 study watersheds in the Kalum area, it is important for all field staff and researchers to follow amphibian hygiene protocols in northwest BC (MOE 2008b).

7. Fringe science: determining the range extent of tailed frogs in northwestern BC (2011-2017). *FLNRO/MOE/UNBC*

A northeast range boundary for the tailed frog was established by Dupuis and Friele (2003) at the north end of the middle-leg of the Zymoetz River (Figure 1). This boundary reflected sampling effort; Dupuis and Friele (2003) sampled to the end of the Kleanza FSR within the Kalum TSA, but no sampling was completed on the upper Zymoetz River past Red Canyon Creek. The boundary of the Coastal Western Hemlock biogeoclimatic zone, formerly assumed to be limiting to tailed frog occupancy, ends just after Mulwein Creek to the east of Red Canyon Creek, and access to the upper Zymoetz is from Smithers, on the Bulkley TSA side.

In 2009, pre-harvest stream assessments for proposed BCTS Babine Business Area harvest blocks confirmed tailed frog adults in two tributaries (one frog in each) in late September (breeding season) flowing into the upper Zymoetz River from the south side (Steventon 2009). No tadpoles were observed. Sites were in the ICHmc1.

To assess factors contributing to Coastal Tailed Frog distributions in this transitional interior area, research sites were established in the upper Zymoetz. In August 2010, as part of the pilot

and site selection project, time-constrained larval searches of streams near the 2009 observations were conducted. To meet the experimental design constraints of the study, stream searches were limited to north-facing streams (located south of the River), within 5km of the confirmed 2009 sightings. Seven streams were surveyed and tailed frogs (adults or larvae) were confirmed in 3 of them; 2 were the original 2009 streams in the Interior Cedar Hemlock (ICHmc1), and one was further east, in the Mountain Hemlock (MHmm1).

To establish tailed frog distribution in the upper Zymoetz, reconnaissance-level, 10-minute timeconstrained searches (2 observers) were conducted in late August at 28 road accessible sites on tributaries to the upper Zymoetz River. The survey focused on tributaries north and south of the River, beginning at the Copper River Bridge crossing (~3 km downstream of McDonnell Lake, headwaters of the river) and extending west for ~15 km along the River and the McDonnell FSR, to where the Mulwein FSR ends at Mulwein Creek.

Tadpoles were detected in 1 new stream located on the north side of the Zymoetz River (south facing) in the ICHmc1, confirming tailed frogs on both sides of the upper Zymoetz. To determine if the reconnaissance-level search effort was insufficient to detect tadpoles at the extremely low densities at which they apparently occur in this area, a sub-set of 7 streams were re-visited in mid-September and intensive, 10 - 1m area-constrained searches were applied. Intensive searches did not improve detection rates; tadpoles were only found in one of the research study streams already known to contain tailed frogs tadpoles.

In 2013, water samples were taken at 12 of the 28 sites surveyed in 2013; 8 of 12 were found to contain tailed frog eDNA (see Project 8 results, below), confirming a broader distribution of tailed frogs throughout the upper Zymoetz than resulted from traditional tadpole search surveys. At low tailed frog densities at the outer edges of the species' range, time- and area-constrained larval searches are not sufficiently sensitive to confirm species' presence and will underestimate distributions.

8. Molecular detection of stream-dwelling amphibians in headwater streams (2012-2014). *FLNRO/UNBC*

Tadpole voucher specimens were collected from research sites in August 2011. In 2012, these specimens were used to develop eDNA marker systems and create positive controls for eDNA analysis (Murray and Flores 2013). Two water sampling protocols (3 – 15ml samples and 1 litre filtered samples) were used to collect water samples from the 15 research sites in two seasons (August and October) in 2012. qPCR performed in an eDNA lab established at UNBC confirmed that water filter samples were more sensitive to eDNA detection than simply collecting vials of water, and the method seemed to be sensitive to larval abundance (i.e., more eDNA detected in high abundance versus low abundance streams (Flores et al. 2013).

To compare the effectiveness of eDNA to conventional time-constrained larval surveys, and to test the sensitivity of eDNA to larval abundance, the 15 research sites and 27 random sites were surveyed and sampled, and an UNBC undergraduate thesis student completed the eDNA analyses. Results confirmed eDNA to be a reliable and effective technique for detecting tailed frog presence, and more sensitive than conventional time-constrained larval surveys (Grob 2013). However, comparisons of eDNA quantity to larval abundance produced inconclusive results. Further research in 2014 will explore factors that affect eDNA concentrations in streams and potentially confound the ability to use eDNA as a predictor of abundance.

9. Genetic connectivity across Coastal Tailed Frog landscapes (2014-2017). UNBC/FLNRO

Starting in 2014, a UNBC-led study of the spatial structure of tailed frog populations begins. A PhD student will examine the coarse- and fine-scale genetic structures of tailed frog populations in the Kalum TSA to assess the geographic scale of genetic differentiation and evaluate if gene flow due to dispersal among populations maintains connectivity within and among streams, watersheds and landscapes (Mosher 2013).

Does the Skeena River pose a barrier to gene flow? Does fragmentation caused by human development (e.g. linear corridors) create genetic barriers? What is the appropriate scale at which to develop conservation strategies?

The student joined research field crews in fall 2013 to familiarize herself with the study area, and will begin field work in 2014.

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Appendix C. 2010-2013 Extension

2010

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2011

- McEwan, A. 2011. Habitat ecology of the post-metamorphic Coastal Tailed Frog. UNBC NRES 705 Course Presentation, 6 December 2011. Prince George, BC.
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2012

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2013

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