DESMAN 2025 PROJECT PROPOSAL

THE COLLARED PIKA:

AN EXPLORATION OF CAPTIVE BREEDING BEFORE A SPECIES REACHES THE BRINK OF EXTINCTION

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Introduction

The Collared Pika (CP; *Ochotona collaris*) is a lagomorph that lives above the tree line in the alpine, subarctic habitat of northwestern North America (Figure 1; Lanier and Hik, 2016). CP demonstrate **metapopulation** dynamics restricted to talus and meadow habitat, with populations going extinct and being recolonized by local dispersal (COSEWIC, 2011). The conservation status of CP varies by region and scale (Table 1), but all assessments describe climate change as an imminent and serious threat to the species (Figure 2).

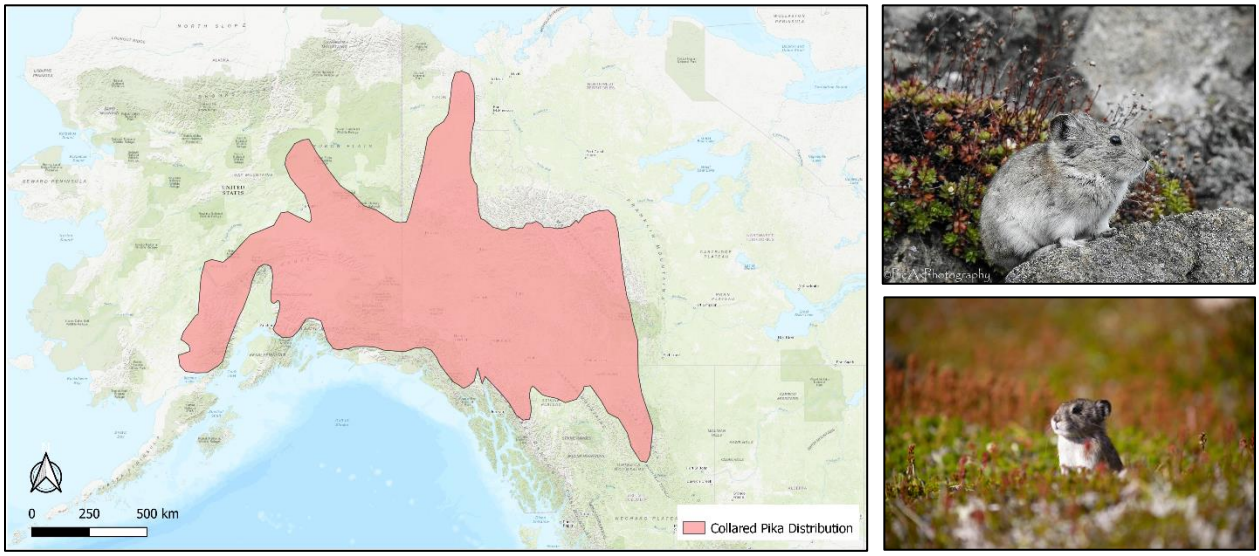

Figure 1. Species distribution map of the Collared Pika (left). Collared Pika occupy talus habitat (top right) and forage in nearby meadows (bottom right). Species distribution data are from IUCN Red List, reproduced on QGIS. Photo credits: Kathy Ax (top right), Mitch Paisker (bottom right).

With their habitat range experiencing climate-shifts in temperature and precipitation faster than anywhere else in Canada (Zhang et al., 2011), it is reasonable to assume that this species is, or soon will be, under much greater threat than its global status currently suggests. With little conservation action able to mitigate the impacts of climate change, captive breeding may be a necessary step in protecting the CP from extinction. Typically, captive breeding is used as a last resort to save species resulting in genetic issues in captive populations (Ralls and Ballou, 2013). By establishing a captive population of CP informed by genetic practices before the species reaches the brink of extinction, we can avoid this issue and effectively protect the CP.

Table 1: Summary of Collared Pika status assessments, by year, across different geographical regions and agencies. All assessments are almost a decade old (see Year Assessed column) and most cite the lack of information available on the species as a cause for concern and a possible source of inaccuracy in determining the status.

Agency	Geographic Region	Status Assigned	Year Assessed
Global Rank			
IUCN	Global	Least Concern	2016
NatureServe	Global	Secure	2017
National Rank			
Species At Risk Act (SARA)	Canada	Special Concern	2017
Committee on the Status of Endangered Wildlife in Canada (COSEWIC)	Canada	Special Concern	2011
NatureServe	Canada	Vulnerable	2016
NatureServe	United States	Secure	2016
Endangered Species Act (ESA)	United States	Not Listed	-
Sub National Rank			
NatureServe	Yukon	Vulnerable; Apparently Secure	2016
NatureServe	Northwest Territories	Vulnerable	2016
NatureServe	British Columbia	Vulnerable; Apparently Secure	2016
NatureServe	Alaska	Vulnerable	2016

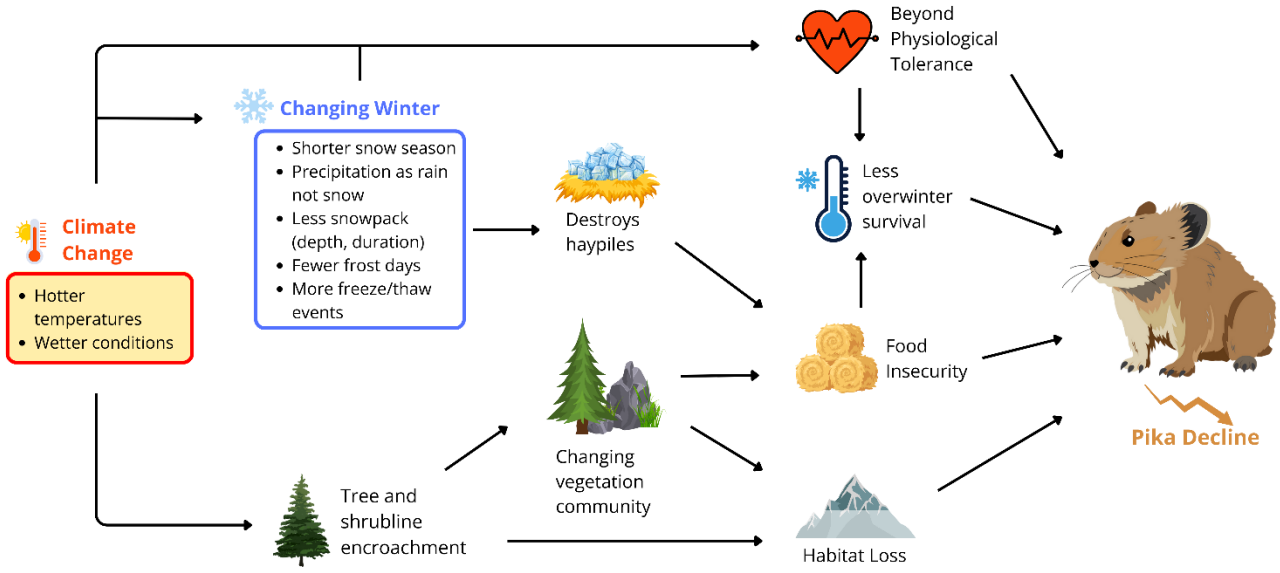

Figure 2: Flow diagram demonstrating the pathways in which climate change is leading to Collared Pika decline by interfering with their physiology and habitat (MacArther and Wang, 1973; Smith, 1974). The changes in winter conditions listed in the blue box cause a greater increase in winter mortality by limiting shelter and thermoregulation benefits of the subnivean environment (COSEWIC, 2011; Morrison and Hik, 2007). Regular freeze/thaw events are delaying the start of the growing season and can cause icing of the Collared Pikas' haypiles

(the vegetation piles they collected and dried during the spring and summer) preventing access to food (Morrison and Hik, 2008). Climate change is also promoting the encroachment of the tree and shrub line further upslope into the Collared Pika's habitat. This encroachment destroys suitable habitat and changes the vegetation community, pushing the Collared Pika further upslope (Lanier and Hik, 2016). The habitat loss caused by climate change is viewed as the Collared Pika's primary threat.

Background

Captive Breeding as a Conservation Strategy

Captive breeding is an "indispensable" conservation tool that aims to maintain a healthy, self-sustaining population of an endangered species (Rahbek, 1993; Ralls and Ballou, 2013). It can be used to create a source population for future reintroductions, provide opportunities for research, and allow for public education and engagement on conservation issues (Frankham et al., 2004; Ralls and Ballou, 2013). With iconic success stories, captive breeding has been credited with saving multiple species from extinction (Bradley et al., 1999; Ralls and Ballou, 2013; Zahler and Rosen, 2013). However, there are several downsides of captive breeding that warrant caution, such as: (1) **inbreeding** and **outbreeding depression**, (2) selection in captivity, (3) loss of learned behavioural traits, (4) animal welfare, and (5) whether the species can successfully breed in captivity at all (Gippoliti, 2004; Ralls and Ballou, 2013). Therefore, to have successful and ethical captive breeding programs, knowledge of species (or analogues), husbandry experience, and genetic planning are required. To safeguard the genetics of captive populations, the best practice is to maintain 90% of genetic diversity of the wild population for 100 years (Miller et al., 2010; Ralls and Ballou, 2013).

Why Captive Breeding is a Good Fit for the Collared Pika

The CP faces a bleak future with climate change continuing to decrease its habitat, increase mortality, and threaten the collapse of metapopulations (Figure 2). With only so much space to move upslope to counteract the effects of climate change, the CP is living on borrowed time. Establishing a captive breeding program can mitigate this risk and provide further opportunities to learn about the species. CPs also possess biological characteristics like limited distribution, low dispersal, and living at the edge of a habitat range which indicate that captive breeding is a worthwhile option (Gippoliti, 2004). At present, there are no known captive populations of this species worldwide.

Box 1: Definitions of Conservation and Genetic Terms

Definitions from *A Primer of Conservation Genetics* by Frankham et al. (2004).

Evolutionary Significant Unit (ESU): Partially genetically differentiated populations that are considered to require management as separate units

F_{IS}: The total inbreeding with within a population due to inbreeding within sub-populations.

F_{ST}: The total inbreeding within a population due to the differentiation among sub-populations relative to the total population.

Inbreeding depression: reduction in reproduction, survival, or other quantitative characters due to inbreeding

Metapopulation: a group of partially isolated populations of the same species that undergo local extinction and recolonizations

Microsatellites: a locus with a short tandem repeat DNA sequence. Microsatellites typically show variable number of repeats and high heterozygosities in populations.

Outbreeding depression: a reduction in reproductive fitness due to crossing of two populations (or sub-species, or species).

Polymerase Chain Reaction (PCR): method used to amplify specific segments of DNA.

Primers: A short nucleotide sequence that pairs with one strand of DNA and provides a free end at which DNA polymerase enzymes begin synthesis of complementary segment of DNA.

Generally, captive breeding is used as a last resort to save species from extinction (Zahler and Rosen, 2013) once it is too late to gather a founding population that can maintain the standard genetic diversity of 90% for 100 years (Ralls and Ballou, 2013). This results in considerable loss of genetic diversity and rapid inbreeding in the captive population that threatens the success of conservation efforts (Frankham et al., 2004). To avoid this issue, the IUCN recommends that captive breeding populations are established before wild populations drop below 1000. Unlike other captive breeding species, the CP has not yet reached the brink of extinction, meaning that a genetically viable captive population could be established. Setting up a captive breeding program before a drastic population decline helps to safeguard the genetics of the species and provides the benefit of time to improve captive breeding methods and fill research gaps before few individuals remain (Figure 3).

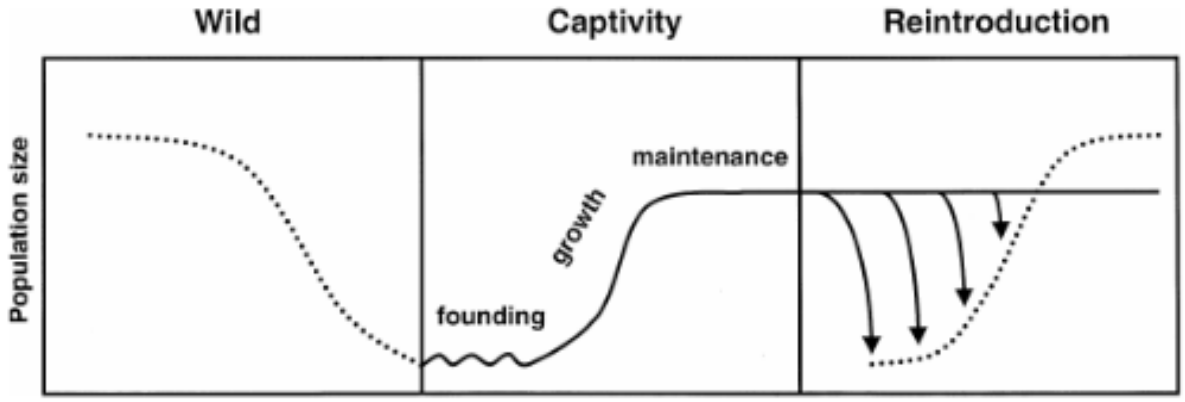

Figure 3: The 6 steps of captive breeding as identified in Frankham et al. (2004) with the dotted line representing the wild population and solid line representing the captive population: (1) the decline of wild populations resulting in loss of genetic diversity and increased inbreeding prior to start of a captive breeding program; (2) founding a small captive population. At this stage, knowledge of husbandry is lacking and a focus on research to improve techniques is needed to ensure reproduction occurs for as many founders as possible; (3) growth stage where the population experiences rapid reproduction and is dispersed across facilities; (4) maintenance phase where the population reaches the target size and does not grow further; (5) Choosing individuals for reintroduction programs; (6) Reintroducing individuals and managing the reintroduced populations.

Setting up a Founding Population of Collared Pikas

The founding stage of a captive breeding program (Figure 3) is arguably the most important step in meeting this goal as it sets the genetic characteristics for the population. To set up a successful founding population one should decide which individuals/populations to collect founders from, collect an appropriate number of founders, and set a proper target captive population size. Theory suggests that the ideal number of founders is 20-30 individuals (Frankham et al., 2004), and for CP, a target population size of ~238 (Box 2).

Box 2: Calculating Target Population Size for the Collared Pika

The target population size needed in captivity to maintain 90% of genetic diversity for 100 years can be calculated as:

$$Ne = \frac{475}{L}$$

where L is equal to generation length (Frankham et al., 2004). For the Collared Pika, the generation time is 2 years (COSEWIC, 2011), meaning Ne ≈ 238 individuals.

The next question concerns where to collect the CP from. Collecting CP from a single population would be the most financially and logistically feasible, but it has its risks. CPs have small populations, fragmented habitat, and low dispersal, which puts them at risk of inbreeding (COSEWIC, 2011). Their metapopulation dynamics also suggest dire inbreeding consequences

caused by frequent bottlenecks from extinction and recolonization events (Frankham et al., 2004). These factors indicate that collecting founders from a single population would create a captive population that is already inbred. However, genetic work on a single metapopulation has shown that CPs are not as prone to inbreeding as theory suggests (Zgurski and Hik, 2014). Overall, there is a knowledge gap as to whether inbreeding or outbreeding depression poses significant risk to a captive CP population making it difficult to determine which populations to collect founders from. It is instrumental to fill this knowledge gap to produce a genetically healthy captive population.

When selecting a population/location, it is also important to consider the guidance of project partners and resources they may have access to. Wildlife Preservation Canada is conservation non-profit with a mission to save animals at risk in Canada, provide opportunities for Canadian biologists to increase their expertise, and advance conservation science with new methods. With experience in captive breeding and a partnership with Greater Vancouver Zoo, Wildlife Preservation Canada is uniquely positioned to develop a captive breeding program for the Collared Pika.

Overall Goal

The development of an effective and ethical captive breeding population of Collared Pika that safeguards the species from extinction in the wild.

Specific Objectives

To identify candidate founder individuals and their populations needed to maintain 90% genetic variation over 100 years, the specific objectives are to:

- Gather **microsatellite** genetic data of potential founders from various populations
- Assess the level of inbreeding in populations to determine whether inbreeding depression would be a concern if all founders were taken from a single population
- Calculate relatedness of sampled individuals to identify non-related candidates for founding individuals

Methodology

Population and Genetic Sampling

The methods for population and genetic sampling are based on Zgurski and Hik (2014). From mid-June to August when CPs are active collecting vegetation, Tomahawk live-capture traps will be placed near CP haypiles and baited with fresh vegetation to catch CPs. A small portion of ear tissue can be taken from captured CPs and should be stored in 95% ethanol at 25℃. Microsatellite DNA samples (see Table 2 for justification) can then be extracted using DNeasy Blood and Tissue Kit and then genotyped with **primers** described in Zgurski et al. (2009). The samples will then need to be amplified using **PCR** for further analysis.

Table 2: The benefits and disadvantages of using microsatellite markers for genetic relatedness analysis (Webster and Reichart, 2005) for objectives 2 & 3. Further context on how the disadvantages do not apply to the Collared Pika are given in sub-bullet points under each disadvantage.

Benefits	Disadvantages

• Easily distinguish heterozygotes and homozygotes allowing precise comparisons between individuals
• High mutation rates in microsatellite markers make it easier to identify related individuals
• Only requires small amounts of DNA
	• Requires species-specific primers in order to amplify DNA with PCRs (Polymerase Chain Reactions)
	  ○ Primers for the Collared Pika already determined by
	• Difficulties in identifying parentage
	  ○ Parentage assignments are not needed to accomplish objectives

Assessing the Risk of Inbreeding Depression within Populations

To determine the level of inbreeding within subpopulations, the SPAGeDi (Spatial Pattern Analysis of Genetic Diversity) program can be used to calculate F statistics $\mathbf{F_{IS}}$ and $\mathbf{F_{ST}}$. Permutation tests can then be used to assess whether the F statistics values are lower (greater inbreeding) than what would be expected under random mating. This will allow us to identify the degree of inbreeding occurring within sampled populations. This analysis is based on methods from Zgurski and Hik (2014).

Producing a Relatedness Matrix of the Sampled Individuals

To identify how related sampled individuals are to each other, a relatedness matrix can be produced using the sampled DNA with the program SPAGeDi or RELATEDNESS. The matrix will present how related each individual is to every other individual. This analysis is based on methods from (McEachern et al., 2007).

Project Schedule

Table 3. Gantt chart showing the overall schedule of activities and tasks undertaken during the 3 stages of this project: pre field season planning, field work, and data analysis and reporting.

	Jan	Feb	Mar	Apr	May	June	Jul	Aug	Sept	Oct	Nov	Dec
Pre Field Season Planning												
Staff Hiring	■											
First aid and other training		■										
Finances & budgeting	■	■										
Permits		■	■	■								
Stakeholder consultations		■	■	■							■	■
Field Work												
Field season logistic planning				■	■							
Purchasing equipment					■							
Field season data collection						■	■	■				
Data Analysis & Reporting												
PCR lab work									■			
Acquire necessary analysis programs									■	■		
Genetic data analysis										■	■	■
Reporting					■			■				■

Expected Outputs

- Relatedness matrix and inbreeding statistics for sampled populations
- A report and peer-reviewed publication on genetic status of CP populations
- Recommendations for population(s) to select founders from, and potential candidates for founders

Expected Outcomes

- Establishing a plan for the start of a captive breeding program for CP run by Wildlife Preservation Canada in collaboration with partners like the Greater Vancouver Zoo
- Raise profile of CP as a conservation target
- Showcase how captive breeding can be used more proactively preventing inbreeding in captive populations

Resources Required

Table 4. Budget breakdown of resources required for Collared Pika project including staffing, field, lab, and contingency costs. All costs are in CAD. Justification and examples/sources for costs chosen are provided.

	Price Per Unit	No. of Units	Cost	Subtotal	Justification	Example/Source
Staffing Costs						
Project Lead Pay	70 000	1	70 000	70 000	Based on reporting for conservation project lead salary in Canada	https://www.glassdoor.ca/Salaries/conservation-program-lead-salary-SRCH_KO0,25.htm
Field Assistant Pay	12 600	1	12 600	82 600	Based on current WPC research assistant posting calculated for summer employment (4 months)	https://wildlifepreservation.ca/wp-content/uploads/2025/03/OPRREC_Research-Assistant-Job-Description-2025_en.pdf
First Aid Training	150	2	300	82 900	First aid training for staff that will be working in remote field conditions	https://www.redcross.ca/training-and-certification/course-descriptions/first-aid-training/wilderness-remote-first-aid-program
Field & Lab Costs						
Tomahawk Trap	110	10	1100	84 000	For trapping Collared Pikas	https://wcscanadastore.com/products/tomahawk-7x7-pro-skunk-trap-with-one-trap-door-and-rear-access-door
Transport	2500	2	5000	89 000	Estimated cost of travel for 3 months of field work	-
Accommodations & Meals	7560	2	15 120	104 120	Based on field station rates in Yukon for non-profit users	https://klrs.ca/wp-content/uploads/2025/04/Kluane_Lake_Research_Station_2025_Rates.pdf
DNAeasy Blood & Tissue Kits	227	3	681	104 801	DNA extraction equipment that can extract 50 samples per kit	https://www.qiagen.com/us/products/discovery-and-translational-research/dna-rna-purification/dna-purification/genomic-dna/dneasy-blood-and-tissue-kit
PCR Runs	90	4	360	105 161	Cost of running PCR analysis	Based on costs identified in (Schlatter et al., 2015)
Contingency Fund						
5 % Contingency	5258	1	5258	110 419	Contingency for unexpected or underestimated funds	-
Total				**$110 419**		

Risks And Assumptions

Table 5. Description of potential assumptions and risks of this project along with the likelihood of them occurring and possible mitigation and response options.

Assumption/Risk	Likelihood	Mitigation/Response
Risk that permits for genetic sampling of Collared Pikas not approved by field season	Medium – permitting can be complicated and subject to bureaucratic delays	Start the permitting process as early as possible to avoid delay; if necessary, delay project until subsequent year
Assumption that all CP populations are the same **evolutionary significant unit (ESU)** and there would not be a *Risk* of outbreeding depression if CPs from different populations were interbred	Low – current genetic work and ecological understanding suggest that all known populations of CP are the same ESU. Newly discovered populations may be a separate ESU based on separate ESUs of the American Pika.	Could use genetic samples gathered to assess whether genetic differentiation between populations is high enough to avoid interbreeding between certain populations.
Assumption that a captive population will be founded within 1-2 years of the study being conducted. CPs live 1-2 years so starting a captive population after this time frame creates the *Risk* that the relatedness matrix cannot be used to identify founder individuals.	Medium-High – this project could be delayed until the captive breeding program is ready to start so founder individuals can be identified during this genetic sampling. Or this project could be done earlier as more of a pilot study. The timelines will depend on multiple factors and partner needs.	If necessary, the identifying founders component of this project (objective 3) can be delayed until right before a captive breeding program can start while the inbreeding analysis (objective 2) can be done earlier as more of a pilot.

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