CITY OF LONG BEACH COYOTE STUDY AND MANAGEMENT PROJECT

A comprehensive study of *Canis latrans* diet through genetic analysis of scat

ABSTRACT

Interactions between humans and local wildlife are inherent to urbanization and have created a demand for management solutions. The Long Beach Coyote Study and Management Program aims to advance the understanding of the urban coyote population in Long Beach, California. In addition to using preexisting data collected by wildlife services, the team is working to assemble more information on the behavior and distribution of urban coyotes by means of scat analysis. The main components of the study include monitoring coyote activity and dispersal patterns, how the urban environment affects coyote living strategies, and a dietary analysis. Now in its second year, this integral part of the project will augment its data through genetic analysis of scat to determine the diet of *Canis latrans*. It is speculated amongst residents that coyotes are the culprits for the loss of their household pets, but genetic analysis of scat samples will determine exactly what the prey items of the Long Beach coyotes are, leading to a more comprehensive understanding of how to coexist with the coyote population.

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Under the mentorship of Dr. Demian Willette

INTRODUCTION

With the rapid urbanization of cities around the United States comes the inevitable interaction between humans and the wildlife of the surrounding area. Looking back as early as the 1980s, human-coyote interaction has been a large area of focus for many urban ecologists (Howell, 1982). Coyote (*Canis latrans*) habitat decrease is clearly an effect of urbanization in Southern California, and in recent years their home ranges have slowly began to decrease and encroach on many residential communities (Figure 1, Riley, 2003. Weckel, 2010). The Long Beach Coyotes Study and Management Program seeks to monitor coyote activity and dispersal patterns, identify how the urban environment affects coyote living strategies, and asses the diet of the various populations. This particular portion of the study and the focus of this proposal is interested in the genetic analysis of coyote scats for use in diet identification, being carried out by myself, Grace Riggs, and fellow student researcher Matthew Sheridan. Coyotes are often blamed for the deaths of pets, so this genetic analysis will serve to pinpoint exactly what the urban coyote diet consists of and therefore how the coyote populations should be managed. This project would serve to answer questions like: What prey items are most present in Long Beach urban coyote diet? Do domesticated pets such as cats and dogs make up a significant proportion of the diet? We hypothesize that domesticated pets do not make up a large portion of urban coyote diet, which is instead significantly comprised of naturally occurring mammals.

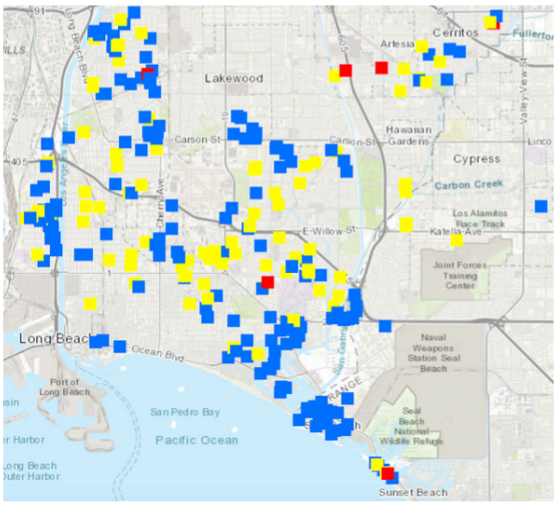

Figure 1: Updated June 2018, this map shows all events in which coyotes were observed in the Long Beach area. Map courtesy of Long Beach Animal Care Services.

- Lowest level incident (siting)
- Moderate level incident (coyote threat)
- High level incident (coyote attack)

BACKGROUND & RELATED WORK

The genetic analysis portion of the Long Beach Coyote Study and Management project is now in its second year. Year one consisted of creating and solidifying methods for Chelex DNA extraction, a simple method that was expected to extract viable DNA for analysis. The first two successful primers were also made during year one and were adjusted and perfected over the course of the last few months to yield the best possible results. The targeted regions were amplified using a process called Polymerase Chain Reaction (PCR). This method allows for selected regions, marked with primers that we created to vary their product size, to be amplified and show us what DNA is present in the samples (Jarman et al., 2004. Symondson, 2002. Espinosa et al., 2015). After around a year of testing, it was determined that there was not enough viable DNA extracted using the Chelex method to be useful in genetic prey identification, so a new method needed to be explored. Scat-specific extraction kits, while costlier, have proven in previous studies to be the most effective way to extract viable DNA from fresh or frozen scat samples. The focus of year two, then, is to utilize and perfect techniques of extraction using stool kits, as well as to create primers to identify more prey species. Currently, the only two primers that are working are for coyote (*Canis latrans*) and domestic cat (*Felis catus*). The next species we would like to be able to identify is domestic dog

(*Canis lupus familiaris),* a feat that is easier said than done because the genomes of domestic dog and coyote are extremely similar in almost every way. When these three primers are run successfully on multiple scat samples, we will move on to making primers for other possible prey species.

We currently have two primers working on our samples (coyote and domestic cat), meaning that we could put these primers in a mix with DNA extracted from scat and run them through PCR and we would see if the scat contained coyote or cat DNA. These two primers were created using the cytochrome B gene (Cyt B) of the mitochondrial DNA (mtDNA). Studies show that between mammals, Cyt B is pretty varied and is therefore fairly easy to build varying primers out of (Bradley & Baker, 2001). While this region worked for creating coyote and cat specific primers, it did not work for domestic dog because this area of the mitochondrial genome in dogs and coyotes is too similar. Because of this, a new region needed to be tested to create viable primers to distinguish domestic dog DNA from coyote DNA, and we believe we have found it in the control region of the mitochondrial genome, which is defined as being between nucleotides 15459 – 16728 (Gundry et al., 2007). This area shows the most promise for creating a primer specific enough to distinguish dog DNA from coyote DNA.

Based on the work completed in year one, the next steps for this project include perfecting DNA extraction techniques using stool-specific extraction kits, creating primers for domestic dog and other prey items, and applying these techniques on a larger scale.

METHODS

Year one consisted of baseline data collection and was predominately focused on perfection of technique and establishment of scientific method. Scats were collected from target sites at varying communities in Long Beach and set aside for DNA extraction. The DNA was extracted using the Chelex method, in which a small amount of scat is put into a tube with Chelex resin and when heated and centrifuged, it traps any possible contaminants which leaves the DNA floating in solution. In the meantime, primers for coyote and domestic cat were successfully created using a series of online tools including NCBI Primer Blast. After several rounds of testing for the presence of viable DNA, it was determined that the Chelex method, while quick and cost-effective, was not adequate for extracting DNA from stool. This leads us to the plan for year two, which will begin with the re-extraction of DNA from the scat samples using stool-specific extraction kits. This method, while a bit more expensive and time consuming, has a very high chance of yielding viable DNA for us to analyze using our primers. In the meantime, the other major focus will be on creation of the domestic dog primer using similar methods as were used for the other primers, but this time looking in the control region of the mtDNA to increase the probability of specificity. Assuming viable DNA is obtained from the stool extraction kits, all of the 35 samples we currently have will be run and categorized based on their contents and

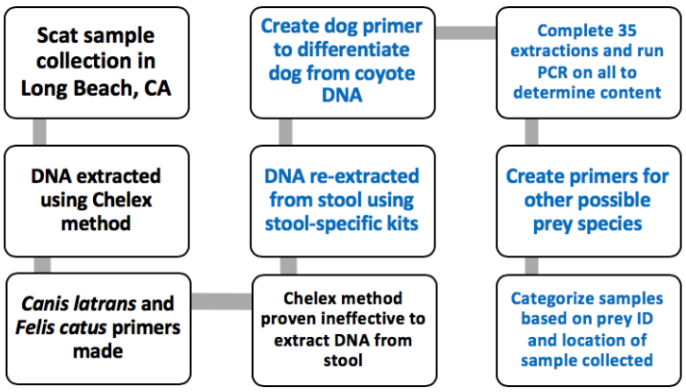

Figure 2: This figure represents the methods from **year one (April 2017 – February 2018)** and the proposed methods for **year two (April 2018 - May 2019)**.

location. The ultimate goal is to be able to run this process in mass quantities so as to be able to analyze urban coyote diet on a larger scale.

EXPECTED RESULTS

The ultimate goal of this project is to provide the City of Long Beach a comprehensive report on how to best manage their urban coyote population. My portion of the project focuses on diet analyzation using genetics, so I hope to be able to contribute an in-depth report on common prey items found in scats from varying areas. The project as a whole will cumulate with publishing a paper and submitting it to the Journal of Urban Ecology and presenting our findings to the City of Long Beach. I hope to be able to confirm our original hypothesis that coyotes are not, in fact, consuming domestic cats and dogs, rather their diet is made up of other more common prey mammals. Besides the actual Long Beach results themselves, I hope to perfect this technique of genetic scat analysis, so it can be used by other researchers on a broader scale to analyze urban coyote diets in other areas.

CONCLUSION

The goal of the City of Long Beach Coyote Study and Management Project is to address citizen concerns and provide a more comprehensive understanding of their urban coyote population. Year one of this study provided a solid footing for establishing more complex and accurate genetic analysis techniques for determining diet, and year two is going to focus on perfecting the techniques and preparing them for use on the larger scale. We hope that in the next year we will be able to provide Long Beach with a comprehensive report on the diet of their urban coyote population by corroborating genetic analysis with game camera footage data. It is our hope that Long Beach will be able to implement our recommended management program in order to assure a peaceful existence between their citizens and their coyote populations.

CITATIONS

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BUDGET

- **Time commitment**
  - Genetic analysis requires a significant amount of time spent in the lab working with precision to produce results that you can present with confidence. Year one required between 4-6 hours in the lab per week, so since year two is ramping up in terms of workload, between **6-8 hours in the lab every week** should be expected in order to work through all the samples in a precise manner.
- **Lab equipment**
  - In terms of lab equipment, Dr. Demian Willette has been generous enough to allow us to use his lab space and equipment to carry out our study, so it would come at no additional cost for me as a student researcher.
  - The QIAGEN QIAmp Fast DNA Stool Mini Kit comes in at **$269.00** for 50 preps. For our immediate purposes, we only need enough to carry out 35 preps for our 35 samples, which means we would be fine with one kit to start out with. However, in the future when we start running more samples it will be necessary to purchase more.