

## Norris Center Natural History Project Award

### Project Description

**Name:** Vanessa Cabrera

**Project Title:** Investigating vector-borne diseases on the University of California, Santa Cruz Forest Ecology Research Plot (FERP)

### Background and justification for project

Zoonotic diseases can place significant risks to human health (e.g. West Nile Virus and Lyme disease). Human habitation and activity in natural areas can increase the incidence of zoonotic spillover events (Gubler 2008). Vector-borne diseases involve complex interactions between host, vector, and pathogen; characterizing the players involved in these disease systems is pivotal for assessing human disease risk.

UC Santa Cruz campus may be a potential hotspot for spillover of zoonotic pathogens to humans as it is bordered by the UC reserve and Wilder Ranch. Little is known regarding the presence (or prevalence) of vector-borne zoonotic diseases at UCSC. We propose to survey small mammal hosts, ticks, and their associated pathogens on the UCSC Forest Ecology Research Plot (FERP). This research will allow us to document vector-borne zoonotic pathogens that naturally occur in this ecosystem.

### Demonstrated initiative and preparation

The Small Mammal Undergraduate Research in the Forest (SMURF) is an undergraduate internship program supporting student research on small mammals. As a SMURF intern, I am directly trained, supervised, and advised by graduate students that have over five years experience working with small mammals and vector-borne pathogens. I am also co-advised by Dr. Beth Shapiro at UCSC and Dr. Andrea Swei at San Francisco State University. Dr. Shapiro teaches an undergraduate course that uses molecular methods to monitor populations of native voles on UCSC reserves. Dr. Swei is an expert on tick-borne pathogens in California (Swei et al. 2010). Additionally, Dr. Swei's lab is equipped with the required facilities and protocols to conduct the molecular identification of tick-borne pathogens.

I have already made significant progress on this project in regards to data collection, and have collected over 60 small mammal tissue samples and over 20 tick samples during fall 2015 and winter 2016 small mammal trapping sessions. I am writing this grant to support further work on analyzing tissue and tick samples for pathogen prevalence.

### Plan for implementation

### Introduction

Preliminary analyses of tick samples from small mammals on the FERP suggest that *Ixodes angustus* may have a unique association with *Peromyscus boylii* (brush mouse) &

*Peromyscus californicus* (California mouse). Interestingly, preliminary data from tick drags did not find any *I. angustus*, and instead indicated a higher abundance of *Ixodes pacificus* on the FERP. We hypothesize that *I. angustus* preferentially parasitize *P. boylii* and *P. californicus*. Previous analyses have also found *Borrelia burgdorferi* present in *I. pacificus* on the FERP (personal communication, Dr. A. Marm Kilpatrick). Although *I. pacificus* is the primary vector for Lyme disease (Brown & Lane 1992), laboratory infection trials have shown that *I. angustus* can also vector the pathogen that causes Lyme disease (Peavey et al. 2000). *B. miyamotoi* is an emerging pathogen in North America, Europe, and Asia, and vectored by *I. pacificus* but current knowledge of its distribution in Santa Cruz County is unknown (Padgett et al. 2014).

We propose a two-step investigation, to examine novel host-vector associations and host-vector-parasite interactions. First, we will investigate host-tick associations in two dominant species of small mammals on the UCSC FERP (*P. boylii* & *P. californicus*). Then, we will molecularly identify two of the most common tick-borne pathogens present in both ticks and the above small mammal hosts, *Borrelia burgdorferi* (agent of Lyme Disease) and *B. miyamotoi*.

### **Questions:**

*Q1. What ticks are present on the FERP, and do they exhibit specific host-parasite relationships?*

*Q2. What biological correlates predict tick burden (e.g. species, sex, etc.)?*

*Q3. What tick-borne pathogens are present in ticks and small mammal species on the FERP?*

### **Methods:**

We will continue to trap small mammals using the same methods used for trapping in fall 2015 and winter 2016. We will use Sherman live traps (2 x 2.5 x 9 inches) to trap *P. boylii* & *P. californicus* over three consecutive nights once a season for spring and summer (2016). Traps will be placed every 20 meters along a 200 x 300 meter grid for a total of 126 traps. We will bait traps with a mixture of oats and peanut butter, and cotton for insulation. We will set traps 1-2 hours prior to sunset and check traps at sunrise the following morning. We will take two 2mm ear punches (n=20 per species) from each collected small mammal, between each animal, ear punch will be flame sterilized. Ear punches will be stored in 2mL tubes filled with 95% ethanol. Feeding ticks will be removed from small mammals using fine-tipped tweezers. Ticks will also be stored in 2mL tubes filled with 95% ethanol. We will conduct tick drags by dragging a corduroy cloth across the forest understory (Swei et al. 2010). Collected ticks will be identified to the species level (Swei et al. 2010). We will extract DNA from ticks and tissue samples using a Qiagen DNeasy extraction kit. We will identify pathogens present in small mammal tissue and ticks using a dual *Borrelia* PCR that will target *B. burgdorferi* and *B. miyamotoi* (Bunikis et al. 2004). Combined, this data will allow us to quantify relative tick species abundance, host-vector associations, and host-vector-pathogen interactions on the FERP.

## Tangible Outcomes

Results from this study will be prepared for publication and submitted as a senior thesis. I will also present this work at conferences, including the UCSC Undergraduate Research Symposium. My research poster will be provided for display at the Norris Center. I will also contribute my findings for the Mobile Ranger natural history tour application.

## Timeline

**Winter 2016:** Small mammal and tick data collection plus tick identification.

**Spring 2016:** Continue with data collection and analyses.

**Summer 2016:** Molecular identification of pathogens (at Dr. Swei's lab) from ticks and tissues.

**Fall 2016:** Write and turn in senior thesis. Produce manuscript for publication.

## Literature Cited

Barbour, A. G., & Fish, D. (1993). The Biological and Social Phenomenon of Lyme Disease. *Science*, 260(5114), 1610–1616.

Brown, R.N., and Lane, R.S. 1992. Lyme Disease in California: A Novel Enzootic Transmission Cycle of *Borrelia burgdorferi*. *Science*, 256 (5062), 1439-1442.

Bunikis, Jonas et al. 2004. Sequence Typing Reveals Extensive Strain Diversity of the Lyme Borreliosis Agents *Borrelia burgdorferi* in North America and *Borrelia afzelii* in Europe. *Microbiology* 150(Pt 6): 1741–55.

Gubler, D.J. 2008. The Global Threat of Emergent/Reemerging Vector-Borne Diseases. Institute of Medicine (US) Forum on Microbial Threats. Vector-Borne Diseases: Understanding the Environmental, Human Health, and Ecological Connections, Workshop Summary. Washington (DC): National Academies Press (US). Vector-Borne Disease Emergence and Resurgence.  
Available from <http://www.ncbi.nlm.nih.gov/books/NBK52945/>

Padgett, Kerry et al. 2014. “Large Scale Spatial Risk and Comparative Prevalence of *Borrelia Miyamotoi* and *Borrelia Burgdorferi* Sensu Lato in *Ixodes Pacificus*” ed. Brian Stevenson. *PloS one* 9(10): e110853.

Peavey, C.A., Lane, R.S., Damrow, T. 2000. Vector competence of *Ixodes angustus* (Acari:Ixodidae) for *Borrelia burgdorferi* sensu stricto. *Experimental & Applied Acarology*, 24(1), 77-84.

Swei, A., Ostfeld, R.S., Lane, R.S, Briggs, C.J., 2010. Effects of an invasive forest pathogen on abundance of tick and other vertebrate hosts in California Lyme disease focus. *Oecologia*, 166(1), 91-100.

Sorensen, T.C., and Moses, R.A. 1998. Host preferences and temporal trends of the tick *Ixodes angustus* in North-Central Alberta. *Journal of Parasitology* 84(5): 902-906

**Budget & Justification**

<b>Item</b>	<b>Cost per Unit</b>	<b>Units</b>	<b>Subtotal</b>
DNeasy Blood and Tissue kit (250 samples)	\$690.00	1	\$690.00
Pipette Tips	\$100.00	3	\$300.00
<b>TOTAL</b>			<b>\$990.00</b>