Lauren Albert
Savaloja Grant Final Report 2018

Project Title: Effect of temperature on the avian immune response to parasitic nest flies

Project Personnel

Lauren Albert
UConn '18

Alexandra Parker
UMN '18

Dr. Sarah Knutie
UConn Assistant

Overview

Hosts have developed defenses to deal with the harm caused by parasites (DeSimone et al. 2017). Resistance as a defense lessens the damage that parasites cause by reducing parasite fitness. The host immune system, such as the antibody response, is an important and well-studied mechanism of resistance. The development of an effective immune response is necessary for host health and survival, and thus can affect the outcome of host-parasite relationships. These host defenses against parasites are affected by changes in the environment. Studying the effect of temperature on host-parasite interactions is important to better understand how these relationships may respond to climate change.

Objectives

The goal of my study is to compare the effect of temperature on the development of an immune response to *P. sialia* in two species of hosts, tree swallows (*Tachycineta bicolor*) and eastern bluebirds (*Sialia sialis*), by manipulating nest temperature and parasite abundance. Tree swallows and eastern bluebirds build their nests in cavities, which include artificial nest boxes. Both species are infested by parasitic nest flies *Protocalliphora sialia* in Minnesota. Adult flies are not parasitic but lay their eggs in the nests of birds. When the fly eggs hatch, the larvae feed on the blood of nestlings and brooding females.

For my study, I made predictions based on the temperature manipulation the nests would receive. For nests in which I increased the temperature, I predicted either no change in eastern

bluebirds or development of an immune response, as higher temperatures will increase speed of development, and either a decrease in immune response in tree swallows due to heat stress, or a strengthened immune response as higher temperatures will increase speed of development (Gillooly et al. 2002, Mashaly et al. 2004). For nestlings in nests that are not infested with parasitic nest flies I predicted there would be no immune response in both eastern bluebirds and tree swallows because the presence of parasites is necessary for nestlings to develop an immune response (Knutie unpublished data).

Photo: Suretypedia

Location

My study was conducted near the Itasca Biological Station, Lake Itasca, MN where Dr. Knutie has studied host-parasite interactions for the past three years, using 170 established nest boxes. The nest boxes have been placed haphazardly on properties of land owners by Dr. Knutie in Hubbard, Beltrami, and Clearwater counties, which surround the biological station (total area: 25km x 35km).

Methods

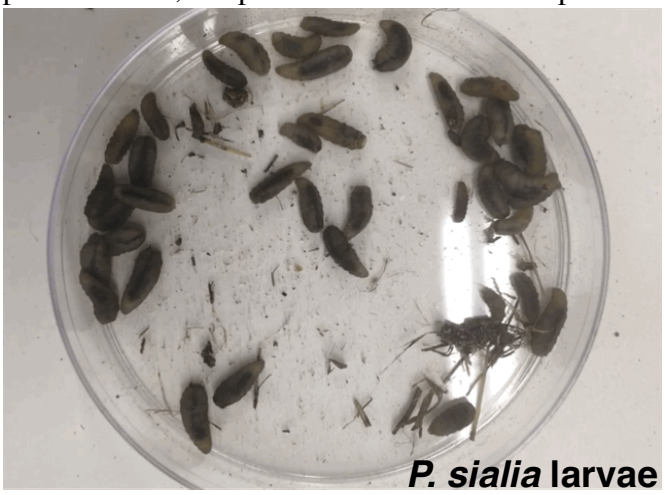
To test effects of temperature and parasite abundance on nestling resistance, I used a 2x2 factorial experiment. The two temperature treatments included: increased and control (no manipulation) temperature. The two parasite treatments included: parasitized or non-parasitized nests. There was a total of 87 nests used in the project for the 2018 field season. Treatment for each species was initially determined by a coin flip and all subsequent nests were alternately assigned to a treatment.

I started temperature manipulation when the first nestlings hatched because King et al. (2010) demonstrated that nestlings begin endogenous synthesis of antibodies from 3-5 days old. I placed an air activated UniHeat 72-hour Heat Packs to manipulate temperature inside the nests assigned to the heat treatment. One pack was placed at the bottom of each nest box and replaced every 2 days. Exhausted packs were used in nests assigned to the no-heat treatment. The target nest temperatures were: heated nests 82°-87°F (28°-31°C and for control nests 70°-75°F (21°-24°C). These target nest temperatures were selected given the average climate data recorded for Lake Itasca, MN (National Centers for Environmental Information 2018). I placed an iButton inside the nest box to record interior temperature.

Nests from non-parasitized treatments were fumigated with a 1% aqueous permethrin solution at hatching as well. Permethrin has been used in previous studies (Knutie et al. 2016) and is harmless to newly hatched nestlings. Nests from parasitized treatments were sham-fumigated

with distilled water. Before treatment, nest contents (nest liner and nestlings) were removed, and then returned once the nest material dried.

When nestlings were 5 and 10 days old they were measured (mass, tarsus, first primary feathers) to quantify growth using a portable scale and dial calipers. Nestlings were uniquely color-banded for easy identification once they began to leave the nest to quantify fledgling success. To prevent forced fledging, I checked nest boxes from a distance with binoculars. I collected a blood sample from 10-day old nestlings to later quantify the immune response. The blood was placed on ice, the plasma and blood was separated at the Itasca Biological Station, and then both samples were placed in a -80°C freezer. The samples were transported to UConn where I began to use an enzyme-linked immunosorbent assay (ELISA) to detect *Protocalliphora*-binding antibodies in the plasma.


***P. sialia* larvae**

After nestlings died or successfully left the nest, nest material was collected and placed in an individual Ziploc bag. The bag of nests were brought to the Itasca Biological Station lab where *P. sialia* larvae, pupae, and pupal cases were be counted to measure parasite abundance.

Progress

I am in the process of completing the enzyme-linked immunosorbent assays using the plasma samples collected from nestlings. The ELISAs are being conducted in the Knutie Lab (Torrey Life Science Building 383) at the University of Connecticut. All nests have been inspected to quantify parasite abundance. Data recorded using the iButtons is also bein analyzed to determine the effectiveness of heat manipulation inside each nest box. Preliminary results for this study were presented at the Frontiers in Undergraduate Research Poster Exhibition at UConn in October 2018. Upon completion of the ELISAs, results will be further analyzed using computer software and a manuscript can be written for publication.

Budget Reconciliation

UConn SURF Grant	$4,000
MOU Savaloja Grant	$1500
UConn EEB Grant	$360
Stipend	-$3000
Total Funding	**$2860**

Itemization:

Housing	$3285
Station Fees	$365
(generously covered by Dr. Knutie)	

Travel

Delta Flight	$485.90

Gas	$618.47
Field Supplies	
Thermocron iButton and Software	$835.95
UniHeat 72 Hour Shipping Warmers	$356.90
Dial Plastic Caliper 150mm	$45.85
Michigan Banding Pliers	$85.00
15cm Wing Rule	$11.00
Cafeteria Tray	$9.95
Velcro circles	$4.88
Purified Water	$4.98
Ziploc Bags	$4.80
Total	$2,494.68
Remaining	$365