

# Molecular Characterization of *Hepatozoon* (Apicomplexa: Adeleorina) Blood Parasite Within the Threatened *Gopherus polyphemus* species

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## Abstract

*Gopherus polyphemus* is a burrowing chelonian endemic to the Southeastern United States. Gopher tortoises are considered a keystone species due to their construction of extensive burrows which provide shelter to approximately 362 commensal species. Haemogregarines are the most common species rich with hemoparasites of reptiles. A previous study has shown that the genus *Hepatozoon* (Apicomplexa: Adeleorina) has been discovered within Jonathan Dickinson State Park gopher tortoise population. We will isolate hemoparasite genomic DNA from blood samples from the gopher tortoises and compare 18S rRNA gene fragment to other *Hepatozoon* species with similar evolution. This project will allow us to better understand the phylogenetic diversity of this parasite in comparison to other *Hepatozoon* species previously discovered.

## Objective

Conduct molecular analysis on the blood of infected gopher tortoise to isolate and identify the 18S rRNA gene & compare to other *Hepatozoon* species with similar phylogeny

## Introduction

### Gopher Tortoise: *Gopherus polyphemus*

- Endemic to SE United States
- Digs extensive burrows
- Keystone species
- **Threatened status**



### Study Site (2014):

- Jonathan Dickinson State Park (JDSP)
- 11,500 acres

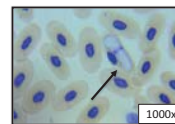


### Haemogregarines

- Most common reptilian hemoparasite
- Three Families
  - *Hepatozoidea*
  - *Karyolysidae*
  - *Haemogregarinidae*

### Genus *Hepatozoon* (1932)

- Potential health effects:
  - Nondetrimental in natural hosts
  - Life threatening in unnatural host



## Method

1) In 2014, blood was collected from three adult gopher tortoises at Jonathan Dickinson State Park.

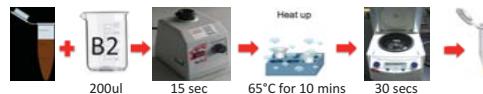
2)

### Genomic DNA Isolation

Step 1:



Step 2:



Step 3:



Step 4:

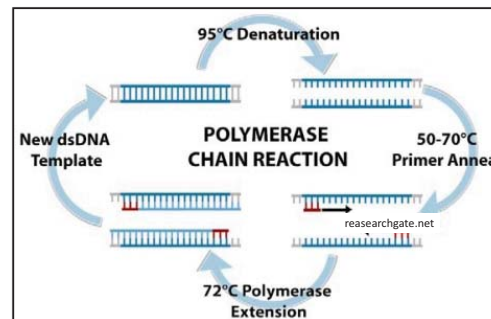


Step 5:



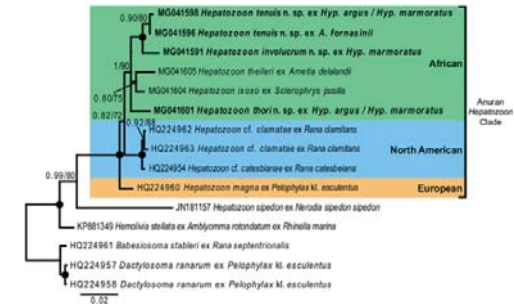
3)

### Polymerase Chain Reaction



## Anticipated Results

### Example of Phylogenetic Tree of *Hepatozoon* sp.



## Future work

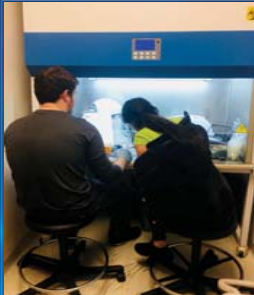
- 1) Order 2 sets of forward and reverse primers to conduct PCR reaction
- 2) Collaborate with Bracken-Grissom laboratory at Florida International University for sequencing and phylogenetic analysis.

## References

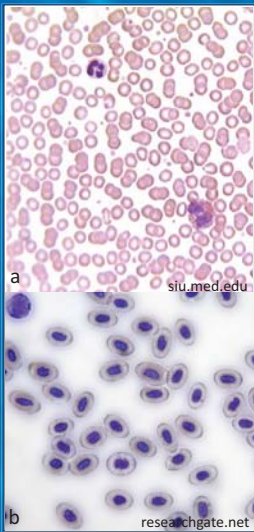
- 1) Harris, D. James, Joao PMC Maia, and Ana Perera. "Molecular characterization of Hepatozoon species in reptiles from the Seychelles." *The Journal of parasitology* 97.1 (2011): 106-110.
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## Acknowledgments

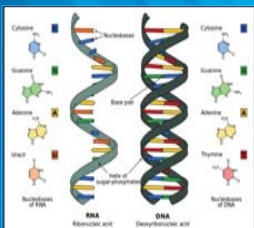
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Isolating DNA from blood samples



Mammalian red blood cells (a) vs Reptilian red blood cells (b)



RNA and DNA strand