DIVERSITY AND PREVALENCE OF *EHRLICHIA* SPP. DETECTED IN IXODID TICKS FROM THE CUMBERLAND GAP REGION OF TENNESSEE, VIRGINIA, AND KENTUCKY.

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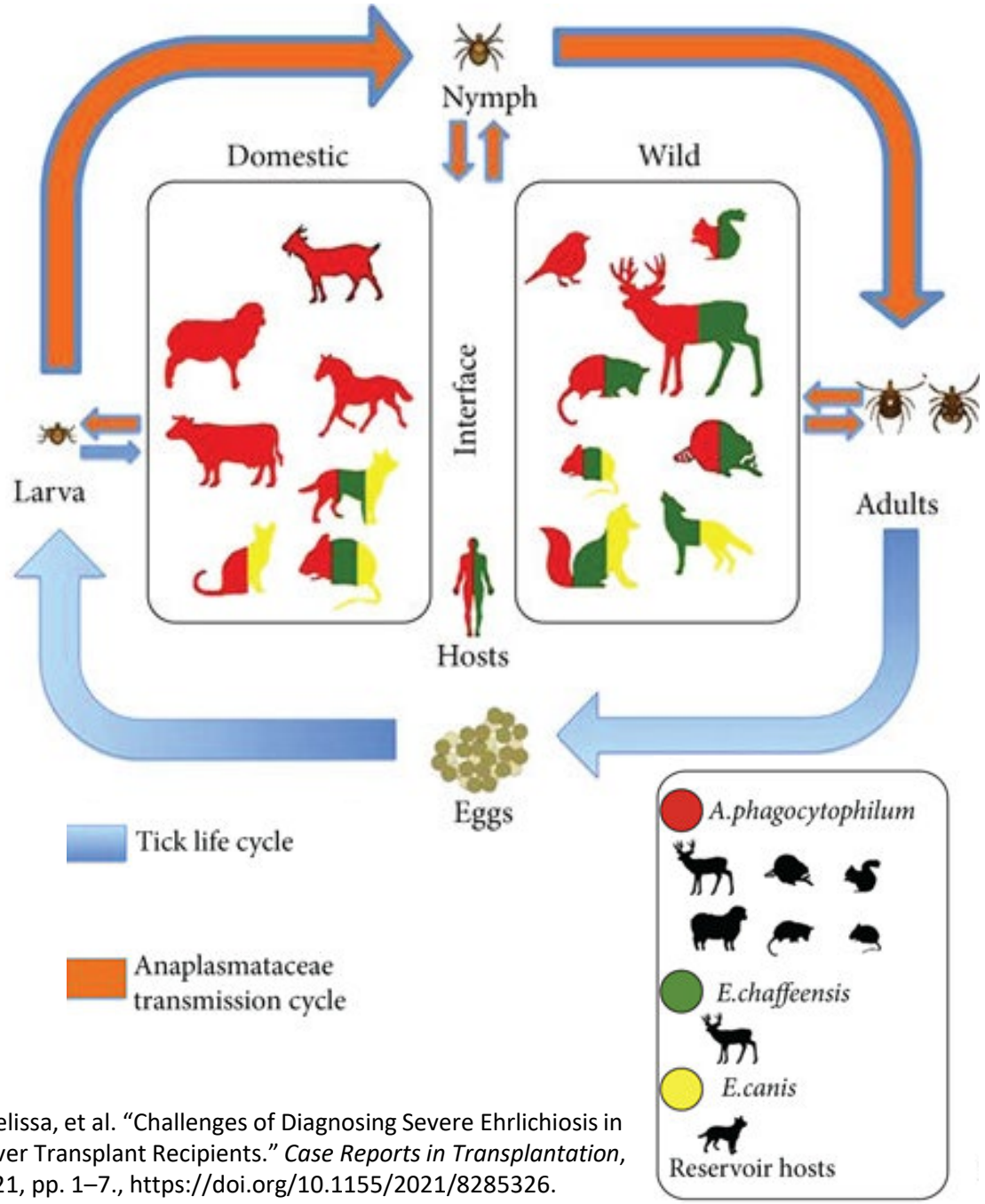
https://www.cdc.gov/ticks/gallery/index.html

EHRLICHIA

- Rickettsiales bacteria in genus Ehrlichia
- Transmitted by lone star ticks (*Amblyomma americanum*) and black-legged ticks (*Ixodes scapularis*)[1]
- Cause Ehrlichiosis in humans, domesticated mammals, and wildlife
- Symptoms include[2,3]:
  - Fever
  - Rash
  - Headache
  - Nausea and vomiting
  - Muscle aches
  - Death



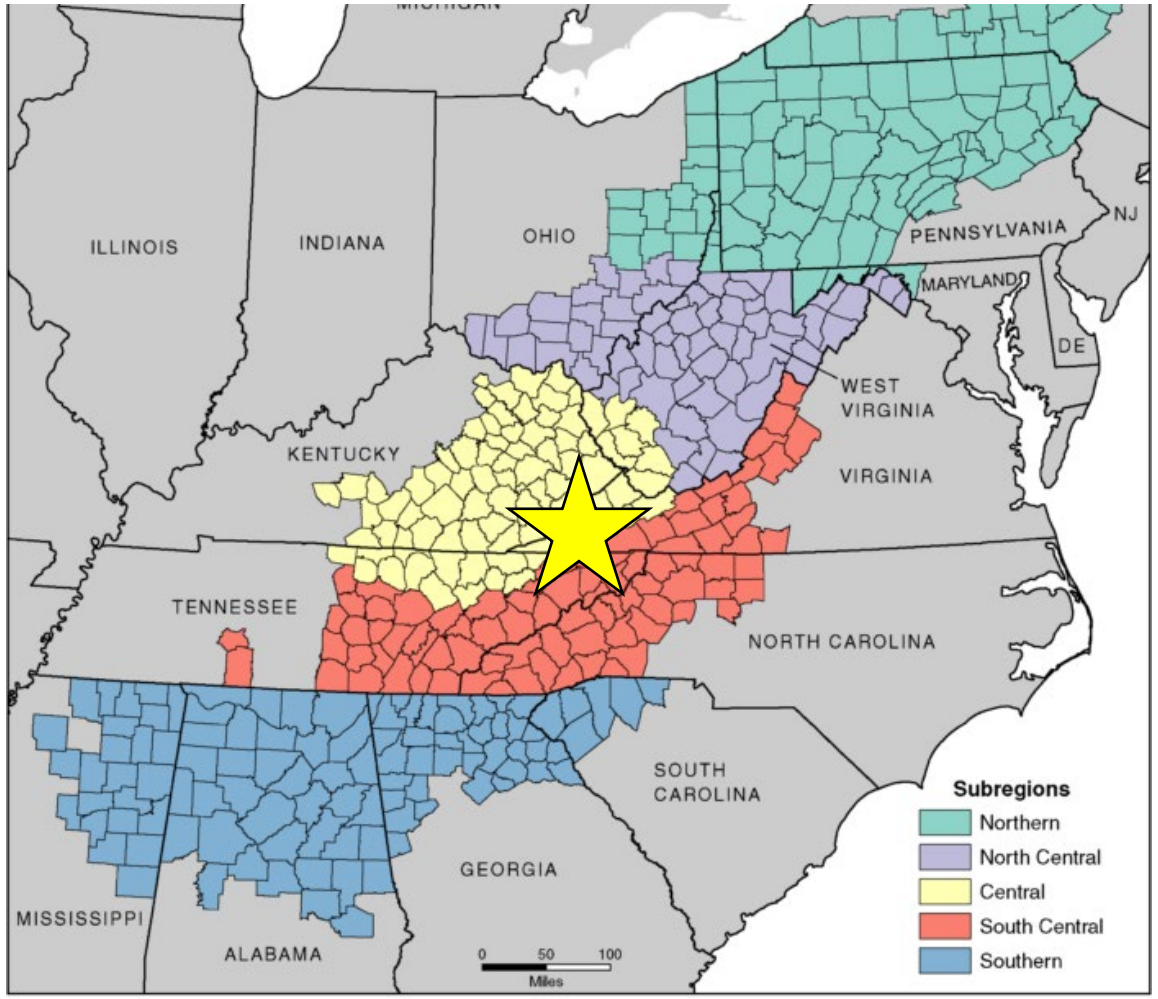
Parkinson, Melissa, et al. "Challenges of Diagnosing Severe Ehrlichiosis in Orthotopic Liver Transplant Recipients." *Case Reports in Transplantation*, vol. 2021, 2021, pp. 1–7., https://doi.org/10.1155/2021/8285326.

RESEARCH QUESTIONS

1. What is the prevalence of *Ehrlichia* species in the Cumberland Gap region?
2. Are *Ehrlichia* species found in tick species other than lone star ticks and black-legged ticks?

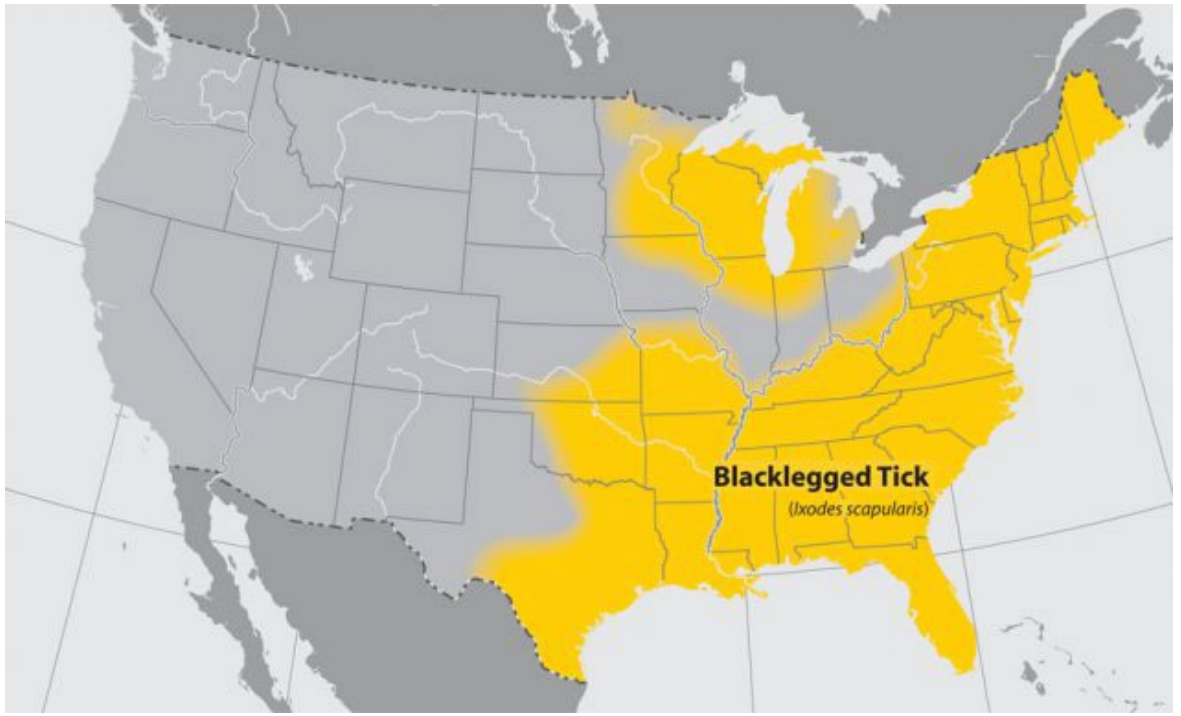
- Hypothesis: We will find a higher prevalence of *Ehrlichia* spp. in lone star and black-legged tick samples than other tick spp. samples.

STUDY AREA



Map by: Appalachian Regional Commission, November 2009.

STUDY AREA



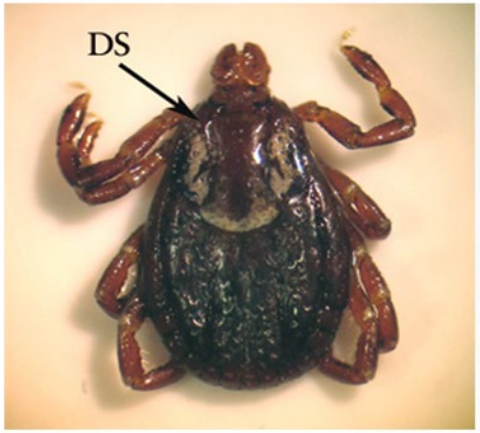
"Transmission." *Centers for Disease Control and Prevention*, Centers for Disease Control and Prevention, 17 Jan. 2019, https://www.cdc.gov/ehrlichiosis/transmission/index.html.

METHODS

1. Identify questing and blood-fed ticks from Tennessee, Kentucky, and Virginia
    - All ticks were collected 2016-2020.
    - Passive collection and collection via tick dragging
    - Ticks identified to species using dichotomous keys
        - Black-legged tick(deer tick), brown dog tick, American dog tick, lone star tick

Female *Ixodes scapularis*
Photo: 2011 Karl Hillig

Female *Rhipicephalus sanguineus*
Photo: CDC

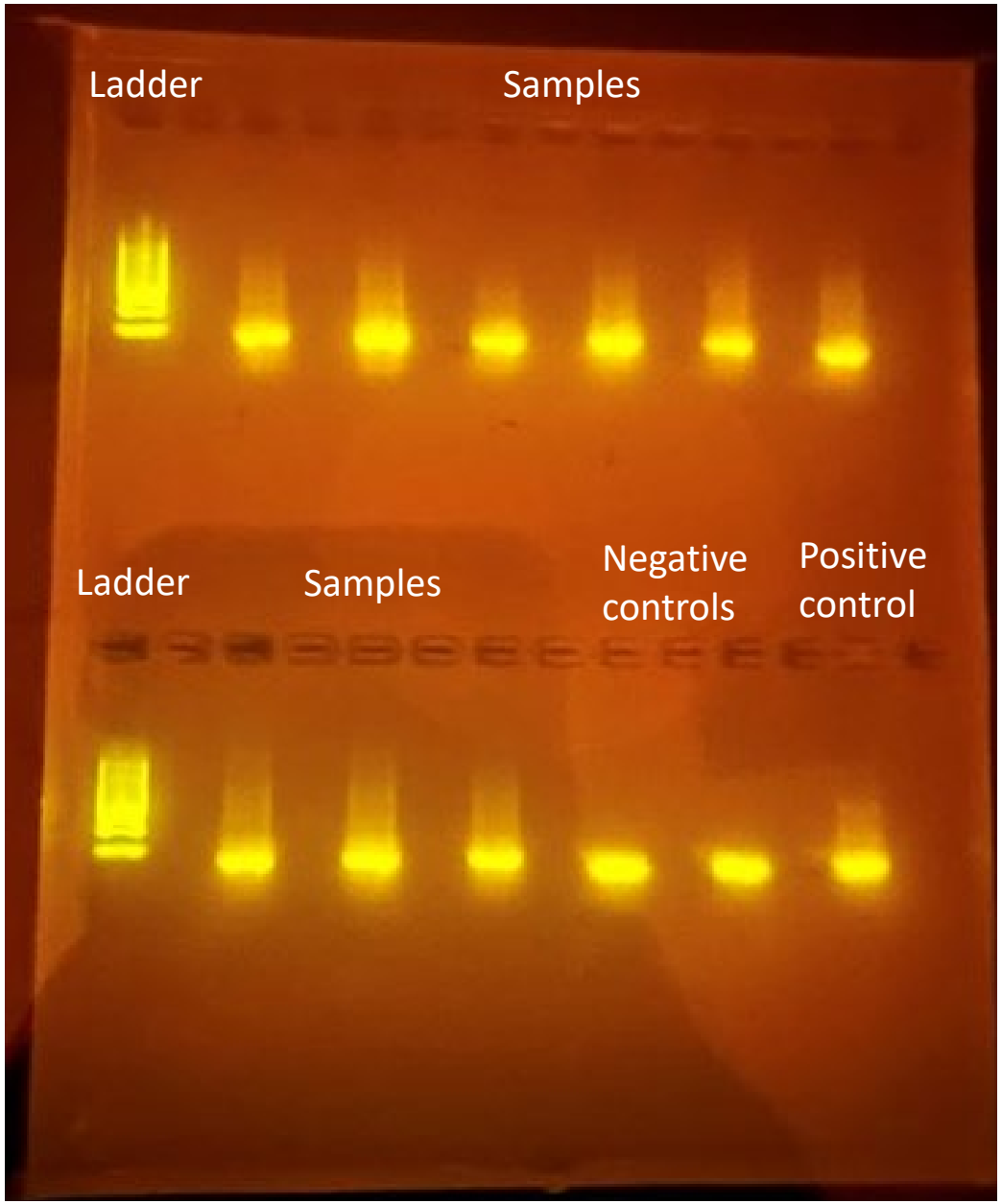


Female *Dermacentor variablis*
Photo: CDC

Female *Amblyomma americanum*
Photo: John R. Maxwell

METHODS

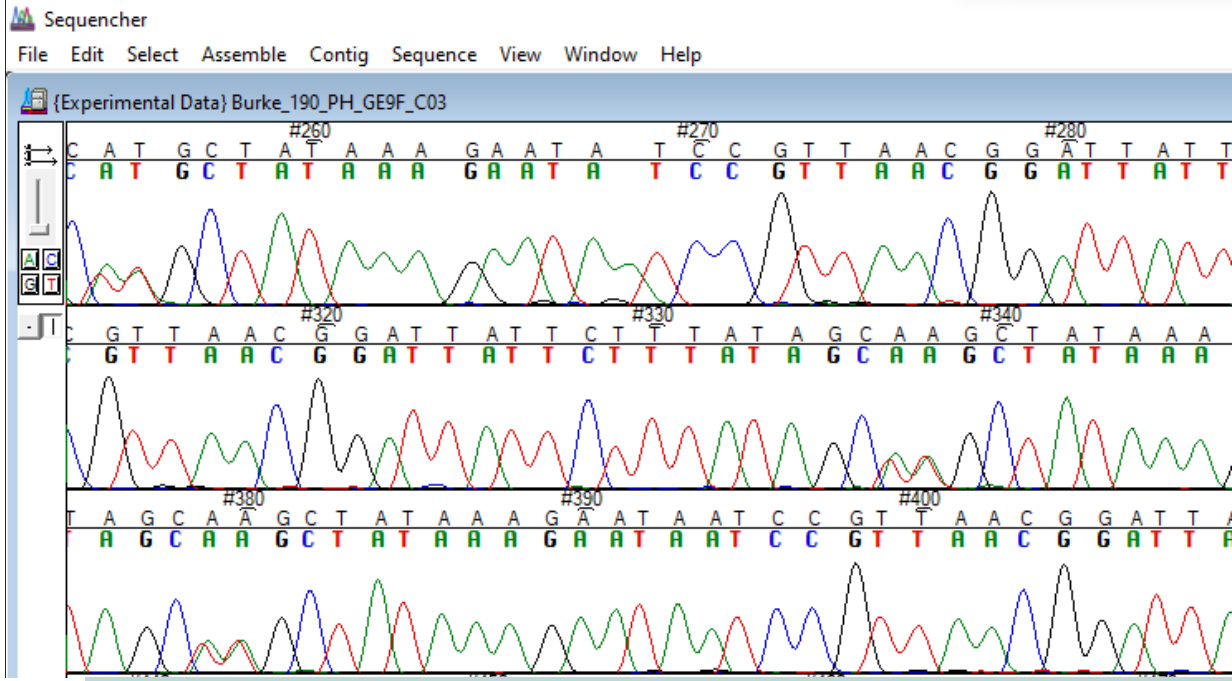
2. Extract DNA from ticks and perform nested polymerase chain reaction (PCR) for all known Ehrlichia spp.
    - Qiagen DNeasy Blood and Tissue Kit (Germantown, MD) used for DNA extraction
    - Gel electrophoresis performed with PCR products
- Goal: 100 samples from each tick species
    - *R. sanguineus, A. americanum, I. scapularis,* and *D. variabilis*

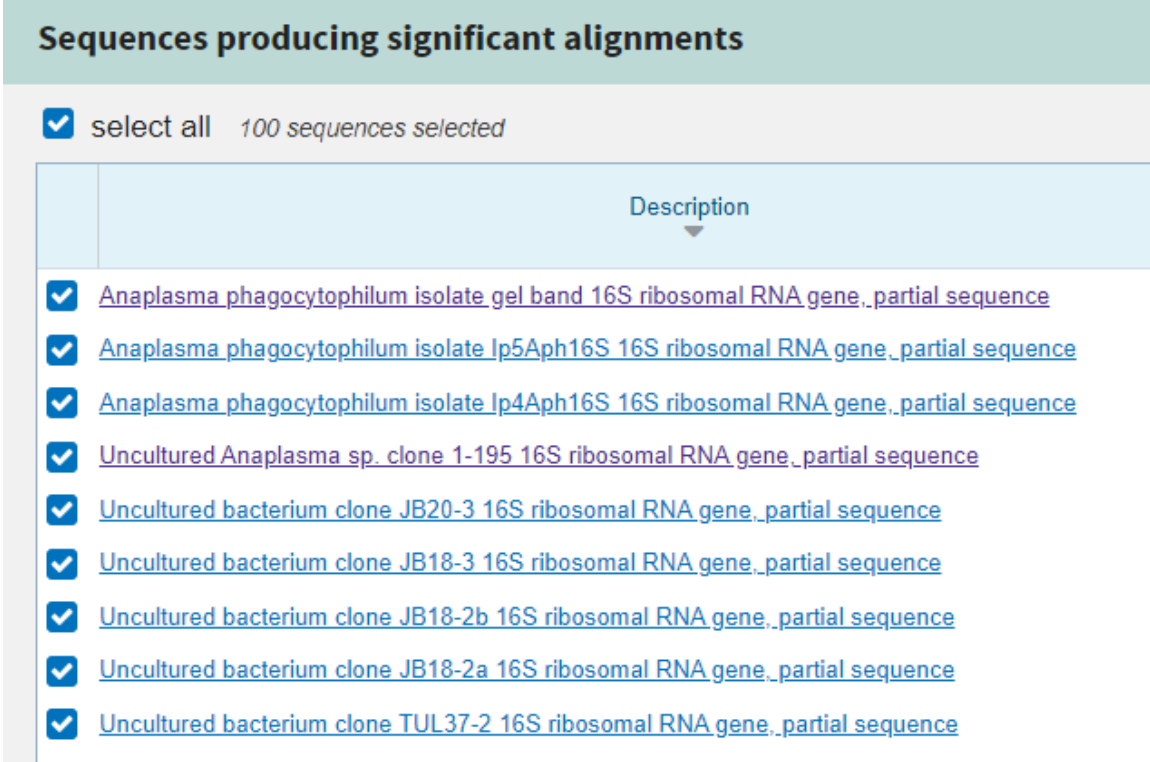


METHODS

3. Sequence positive amplicons to determine which *Ehrlichia* spp. present.

- McLab DNA Sequencing
- Sequencher program
- NCBI Basic Local Alignment Search Tool (BLAST)





RESULTS

- So far, have detected 7 *Ehrlichia* positive and 23 *Anaplasma* positive tick samples
  - *Ehrlichia*: 6 from lone star ticks, 1 from black-legged (deer) ticks
  - *Anaplasma*: 19 from lone star ticks, 4 from black-legged (deer) ticks
- Confirmed via DNA sequencing
- Some other bacterial spp. identified
  - *Coxiella*

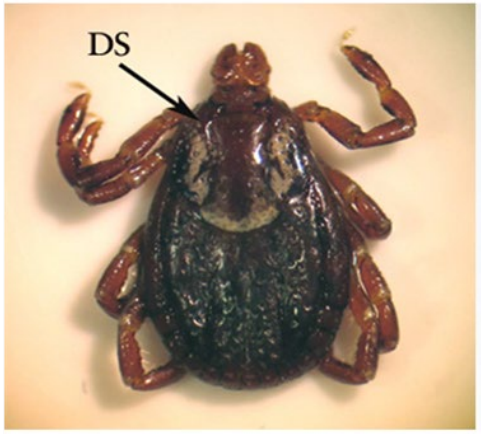
Sample Name	Primary/ Secondary PCR	Gel Positive(s)	Sequencing results
AM 70	primary	Ehrlichia (general)	
Burke 141	primary	Ehrlichia (general)	Ehrlichia (general), A. phagocytophilum
Burke 142	primary	Ehrlichia (general)	E. chaffeensis, A. Phagocytophilum
AM 46	secondary	EW	
Burke 141	secondary	PL, PH	Ehrlichia (general), A. phagocytophilum
Burke 142	secondary	PL, CA, EW, PH	E. chaffeensis, A. Phagocytophilum

FUTURE DIRECTIONS

- Finish PCR for *R. sanguineus* and *D. variabilis*
- Send primary and secondary positives out for sequencing
- Send off all positives for a second round of sequencing to ensure accurate results
- Run statistical tests to see if there is significant correlation between tick spp. and *Ehrlichia* spp.
- May test different primers to see if more specificity

Female *Ixodes scapularis*
Photo: 2011 Karl Hillig

Female *Rhipicephalus sanguineus*
Photo: CDC



Female *Dermacentor variablis*
Photo: CDC

Female *Amblyomma americanum*
Photo: John R. Maxwell

IMPORTANCE

- Further understanding of tickborne diseases
- Recent increase in tickborne disease prevalence in the U.S.[4]
- Lack of tick research in Appalachia despite high tick presence
- Improve knowledge of *Ehrlichia* transmission for doctors and veterinarians

SOURCES

1. Beall, Melissa J, et al. “Seroprevalence of Ehrlichia Canis, Ehrlichia Chaffeensis and Ehrlichia Ewingii in Dogs in North America.” *Parasites & Vectors*, vol. 5, no. 1, 2012, https://doi.org/10.1186/1756-3305-5-29.
2. Fang, Quentin Q., et al. “Prevalence of the Agent of Human Granulocytic Ehrlichiosis in *Ixodes Scapularis* (Acari: Ixodidae) in the Coastal Southeastern United States.” *Journal of Medical Entomology*, vol. 39, no. 2, 2002, pp. 251–255., https://doi.org/10.1603/0022-2585-39.2.251.
3. “Signs and Symptoms.” *Centers for Disease Control and Prevention*, Centers for Disease Control and Prevention, 17 Jan. 2019, https://www.cdc.gov/ehrlichiosis/symptoms/index.html.
4. André, M.R., 2018. Diversity of Anaplasma and Ehrlichia/Neoehrlichia Agents in Terrestrial Wild Carnivores Worldwide: Implications for Human and Domestic Animal Health and Wildlife Conservation. *Frontiers in Veterinary Science*, 5.
5. Parkinson, Melissa, et al. “Challenges of Diagnosing Severe Ehrlichiosis in Orthotopic Liver Transplant Recipients.” *Case Reports in Transplantation*, vol. 2021, 2021, pp. 1–7., https://doi.org/10.1155/2021/8285326.

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QUESTIONS?


Tick season is here
Will hitchhike for food
Brianna Cooper 2014
Lyme Memes For Education