Evaluating wildlife/tick systems in Tennessee for maintenance of the Lyme disease pathogen, *Borrelia burgdorferi*

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OUTLINE

I. Introduction and Background:
   A. Ticks species in Tennessee
   B. Lyme Disease: Ecology
   C. Lyme Disease: United States

II. Objectives and Justification

III. Methods

IV. Preliminary Results

Introduction: Ticks

Class: Arachnida
Order: Ixodida
Family: Ixodidae
Ticks in Tennessee

The only tick species in the eastern U.S. capable of transmitting the Lyme Disease pathogen to humans

Dermacentor variabilis
American Dog Tick

Dermacentor albipictus
Winter Tick

Amblyomma americanum
Lone Star Tick

Amblyomma maculatum
Gulf Coast Tick

Ixodes scapularis
Black-legged Tick (= ‘Deer Tick’)

Introduction: Lyme Disease

Pathogen: Borrelia burgdorferi

Vector: Ixodes scapularis

Reservoir Host: Peromyscus leucopus (white-footed mouse)

Definitive Host: Odocoiles virginianus (white-tailed deer)

Lyme Disease is mainly transmitted to humans by nymphal I. scapularis ticks
(Larvae aren’t infected; adults are easier to see and remove)
1996: CDC map of tick species that spread Lyme Disease

- In the west: *Ixodes pacificus*
- In the east: *Ixodes scapularis*

2003 - 2007: New surveys to undertaken to update the 1996 CDC tick map.

- 96 sites sampled
- State parks and other public land
- 4-6 visits during summer

The Lyme Disease Risk Map Consortium:
Michigan State University, Yale University, University of Illinois

My Objectives

**Goal:** to gain a better understanding of the human risk associated with the blacklegged tick in Tennessee, and of the role wildlife play in perpetuating this risk.

1. To determine the current distribution of *I. scapularis* in TN.
2. To determine the seasonal phenology (life cycle/activity) of *I. scapularis* in TN.
3. To assess host preference (mice vs. lizards) of *I. scapularis* in the state.
4. To determine if *I. scapularis* in TN are infected with *Borrelia burgdorferi* and/or other zoonotic pathogens.
Are we at risk of Lyme Disease in Tennessee?

It has been **anecdotally** claimed that:

- Blacklegged ticks are rare in Tennessee (?)
- The seasonal life cycle of *I. scapularis* in the South ‘breaks’ the Lyme disease transmission cycle (?)
- Lizards don’t support the bacteria, and are the preferred host for nymphal *I. scapularis* in Southern states (?)
- Nymphs’ preference for lizards means that mammals (including humans) are less likely to be bitten (?)

However, there is little or no published evidence for these claims.

Evidence for these claims?

I. *“Blacklegged ticks are rare in Tennessee”*

Prior to 2006, there were only 8 county records of *I. scapularis* in the state (CDC, 1996)

Evidence for these claims?

II. *“The seasonal life cycle of *I. scapularis* in the South breaks the Lyme disease transmission cycle”*
### I. scapularis life-cycle

2-year cycle in northern states
1-year cycle in southern states (?)

- **Adults**
  - egg-lay

- **Larvae**
  - molt

- **Nymphs**
  - molt

- **Third host**
  - Feed + mate

- **Second host**
  - Feed

- **First host**
  - Feed

Source: L. Beati, US National Tick Collection

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**During northern summers, the nymphs are active before the larvae**

This allows the nymphs to infect mice that the larvae can later feed on
(Larvae are uninfected at birth)

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**During southern summers, larvae precede the nymphs, perhaps ‘breaking’ the disease cycle?**

Seasonal timing of black-legged tick life-stages in South Carolina
III. “Nymphs' preference for lizards in the South lowers the risk that mammals (including humans) will be bitten”

“Lizards don’t support the bacteria, and are the preferred host for nymphal I. scapularis in Southern states”

Evidence for these claims?

Immature I. scapularis are found on at least 14 species of lizards (Keirans et al. 1996)

Since lizards do not support the Lyme Disease spirochete, Fish and Howard (1990) argue that the risk of Lyme Disease is low in Southeastern states, where numerous lizard species are available as hosts for I. scapularis ticks
Methods to investigate these claims

**Field**

1. Deer Check-station Survey for distribution
2. Vegetation ‘Drag’ Surveys for seasonal phenology
3. Mammal and Lizard Trapping for host preference

**Laboratory**

1. Identify all ticks
2. Quantify tick engorgement
3. Extract total tick DNA, amplify, and visualize potential *Borrelia burgdorferi* DNA
4. Purify and sequence positive samples

Field: Deer Check-station Surveys

Counties where we checked deer for ticks in 2007

Field: Monthly Surveys

Henry Horton State Park
Field: Vegetation ‘Dragging’

- 1m X 1m white corduroy cloth
- 100 meter transects
- 5 transects per site
- Transect evenly distributed in site

Field: Small Mammal and Lizard Trapping

- Sites sampled monthly for at least 1 year
- 3 Transects per site
- 25 Sherman live-traps per transect
- Traps set 10 m apart
- Pitfall & funnel traps for lizards
- Ticks, tissue, blood samples

Weather Measurements:
- Temperature
- Barometric Pressure
- Relative Humidity

Lab Methods

Identify
- All ticks to species
- Separate each species & life stage

Measure
- Scutum Width (SW)
- Body Length (BL)
- Engorgement Index
  \[ SW / BL \] (Falco et al. 1996)
Lab: Extract. Amplify. Visualize & Sequence Pathogen DNA

**Extraction:** Qiagen DNEasy Blood & Tissue Kit

**Amplify:** PCR (Tsao et al, 2004)

**Visualize:** Agarose Gel

**Purify:** Promega DNA Purification

**Sequence:** To determine species and strain

Preliminary Results: Deer Check

<table>
<thead>
<tr>
<th>Species</th>
<th>Adult Female</th>
<th>Adult Male</th>
<th>Nymph</th>
<th>Larvae</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>I. scapularis</em></td>
<td>243</td>
<td>183</td>
<td>0</td>
<td>0</td>
<td>426</td>
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<tr>
<td><em>D. albipictus</em></td>
<td>483</td>
<td>797</td>
<td>706</td>
<td>11</td>
<td>1997</td>
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<tr>
<td><em>A. americanum</em></td>
<td>3</td>
<td>31</td>
<td>1</td>
<td>2</td>
<td>37</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>729</strong></td>
<td><strong>1011</strong></td>
<td><strong>707</strong></td>
<td><strong>13</strong></td>
<td><strong>2460</strong></td>
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</table>

<table>
<thead>
<tr>
<th>Region</th>
<th>Deer Checked</th>
<th>Deer with <em>I. scapularis</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Region 1</td>
<td>71</td>
<td>3</td>
</tr>
<tr>
<td>Region 2</td>
<td>42</td>
<td>24</td>
</tr>
<tr>
<td>Region 3</td>
<td>130</td>
<td>30</td>
</tr>
<tr>
<td>Region 4</td>
<td>363</td>
<td>9</td>
</tr>
<tr>
<td>Oak Ridge</td>
<td>74</td>
<td>7</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>680</strong></td>
<td><strong>73</strong></td>
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Tennessee Counties with *Ixodes scapularis*
**Preliminary Results: Deer Check**

**Tennessee Counties with *Ixodes scapularis***

- Major Tennessee Cities
- Records prior to 2006
- New records in 2006

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**Preliminary Results: Deer Check**

**Tennessee Counties with *Ixodes scapularis***

- Major Tennessee Cities
- Records prior to 2006
- New records in 2006
- New records in 2007

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**Our findings so far…**

- Blacklegged ticks are rare in Tennessee?
  - They are relatively common in many counties …
- The seasonal life cycle of *I. scapularis* in the South ‘breaks’ the Lyme disease transmission cycle?
  - We have found nymphs active in February…
- Nymphs’ preference for lizards in the south lowers the risk that mammals (including humans) will be bitten?
  - We don’t know yet - however nymphs are definitely ‘draggable’ at HHSP …
Are *I. scapularis* ticks in TN infected with the *Borrelia burgdorferi* spirochete – and/or other zoonotic pathogens?

We will know soon!

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Questions?
Deer Check-station Results 2007

Proportion of deer by county carrying *I. scapularis* ticks

- **0 %**
- **1 – 24 %**
- **25 – 100 %**

Solid shading indicates 5+ deer checked

Diagonal shading indicates 1-5 deer checked