UNIVERSITY OF MOUNT UNION
HUSTON-BRUMBAUGH
NATURE CENTER

BRUMBAUGH Bytes

Do Ticks Carry More Than One Passenger? A Story of Co-Infection in the Forest at Huston-Brumbaugh Nature Center (HBNC)

Featured scientist: Emily Krizner, University of Mount Union, Brumbaugh Scholars Program

Research Background:

Emily Krizner is fascinated by tiny creatures that can carry big consequences—ticks. Specifically, she studies the blacklegged tick, *Ixodes scapularis*, which is known for spreading **Lyme disease** through a bacterium called *Borrelia burgdorferi*. Young (nymph) and adult blacklegged ticks (also called deer ticks) are the main life cycle stages that spread Lyme disease. These ticks like to live in forests or places with lots of plants. You'll often find them in the same areas where white-tailed deer live, because deer help ticks move around and find new places to live. But Emily wasn't just interested in Lyme disease. She wanted to know: **What happens when ticks carry more than one kind of bacteria?**

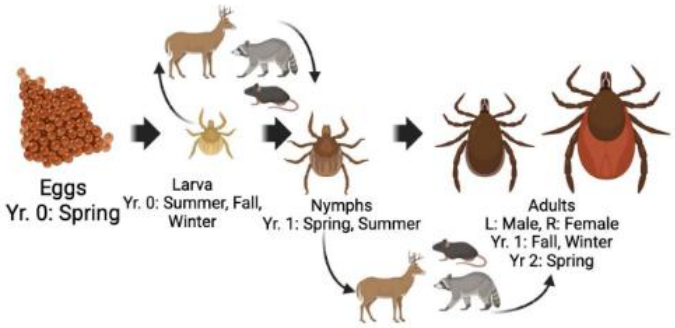

Figure 1. *Ixodes* scapularis Life Cycle. Created in BioRender.com
Adapted from: "Blacklegged tick (*Ixodes scapularis*) lifecycle" (CDC 2024)

One of those bacteria is *Wolbachia*, a microbe that lives inside the gut of many insects. In mosquitoes, *Wolbachia* can actually help stop the spread of diseases. It does this in a few clever ways. For example, it can change how mosquito genes work, it can turn male mosquitoes into females, and it can even block viruses from growing inside the mosquito's gut. All of these tricks help prevent viruses—like dengue or Zika—from being passed on to humans. But what about in ticks? Could *Wolbachia* affect how likely a tick is to carry the Lyme-disease causing bacterium *Borrelia burgdorferi*?

Emily wanted to identify whether Wolbachia and B. burgdorferi displayed coinfection or competition in Ixodes scapularis at the Huston-Brumbaugh Nature Center (HBNC).

1. What do you already know about Lyme disease and how it spreads?

   _____

   _____

2. What do you think might happen if a tick carries both *Wolbachia* and *Borrelia*?

   _____

   _____

Research Questions:

3. Use a highlighter to identify the research question Emily wanted to answer with her research.

Created by Amy Miller
HBNC Environmental Education Outreach Coordinator
Content developed with support from Microsoft Copilot

***Hypothesis*:**

4. Write a **null hypothesis** for this experiment?

_____

_____

_____

_____

5. Write an **alternative hypothesis** for this experiment?

_____

_____

_____

_____

Methods:

Emily collected 40 tick nymphs from an old-growth forest at the HBNC between May and August 2025. She used a DNA extraction kit to isolate genetic material from each tick. Then she used PCR (Polymerase Chain Reaction) to test the three genes:

- CO1 – a gene found in all arthropods (used to confirm the DNA extraction worked)
- 16S rRNA – a gene specific to Wolbachia bacterium
- OspA – a gene specific to Borrelia burgdorferi bacterium

She used gel electrophoresis to visualize the results and determine which ticks carried which bacteria.

6. Why do you think Emily tested for the CO1 gene in every tick?

_____

_____

_____

7. What does it mean if a tick is positive for 16S rRNA or OspA?

_____

_____

_____

Created by Amy Miller
HBNC Environmental Education Outreach Coordinator
Content developed with support from Microsoft Copilot

Scientific Data:

Here's what Emily found:

Table 1. CO1, 16S rRNA and OspA Results in surveyed I. scapularis nymphs

Gene Tested	Number of Positive Ticks (out of 40)
CO1	37
16S rRNA (Wolbachia)	17
OspA (Borrelia)	4

8. Create a bar graph displaying this data. Be sure to include a Graph Title and label the axes.

Table 2. Among the 37 ticks with conclusive results:

- 14 had Wolbachia only
- 3 had both Wolbachia and Borrelia
- 1 had Borrelia only
- 19 had neither

9. Create a bar graph displaying this data. Be sure to include a Graph Title and label the axes
.

Created by Amy Miller
HBNC Environmental Education Outreach Coordinator
Content developed with support from Microsoft Copilot

10. Based on this data, do you think *Wolbachia* prevents *Borrelia* infection in ticks? Why or why not?

_____

_____

_____

_____

Interpretation:

Emily found that co-infection does occur—some ticks carried both bacteria. However, there was no clear pattern showing that *Wolbachia* prevents *Borrelia* infection. In fact, most ticks didn't carry *Borrelia* at all, and nearly half carried *Wolbachia*.

11. What conclusions can you draw from Emily's data?

_____

_____

_____

_____

12. What would you want to test next if you were continuing this research?

_____

_____

_____

_____

Limitations:

Emily noted a few challenges:

- She only sampled ticks from one part of the forest.
- The OspA gene might not always be detectable if the infection is inactive.
- She only tested for one *Borrelia* gene—others might give more accurate results.

13. How might these limitations affect the results?

_____

_____

_____

_____

Created by Amy Miller
HBNC Environmental Education Outreach Coordinator
Content developed with support from Microsoft Copilot

14. What could Emily do differently in a future study?

_____

_____

_____

_____

_____

Try It Yourself!

15. Imagine you're Emily. You've just collected 50 more ticks. Design a follow-up experiment to test whether *Wolbachia* affects *Borrelia* infection. What would you change or add?

_____

_____

_____

_____

_____

_____

_____

_____

_____

_____

_____

_____

_____

_____

_____

_____

_____

_____

_____

Citations:

1. **Krizner, E. (2025).** *Investigating Co-Infection and Competition between Wolbachia and Borrelia burdorferi in Ixodes scapularis.* Brumbaugh Scholars Program, University of Mount Union, Alliance, OH 44601