Cold Tolerance and *Bartonella* Infection in *Dermacentor Variabilis* (American Dog Ticks) From Nova Scotia

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Background:

Ticks are important parasites and vectors of bacterial pathogens that affect human and animal health. In Nova Scotia, the American dog tick, *Dermacentor variabilis*, is common in disturbed and edge habitats and can carry *Bartonella spp*., a group of bacteria associated with a range of clinical signs in humans and domestic animals.

Overwinter survival is a key bottleneck for tick populations. Cold tolerance traits such as critical thermal minimum (CTmin), chill-coma recovery time (CCR) and supercooling point (SCP) describe:

- How cold a tick can go before losing coordination (CTmin)
- How quickly it recovers from chill coma (CCR)
- The temperature at which its body fluids freeze (SCP).

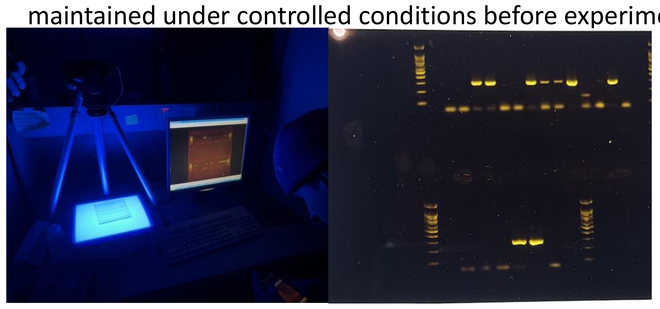
Although infection can influence energy reserves and physiology in arthropod vectors, the association between Bartonella infection and cold tolerance has not been examined in Nova Scotian D. variabilis.

Figure 4: Study organism American dog tick (*Dermacentor variabilis*), dorsal view. *D. variabilis* is common in Nova Scotia and was used here to test whether cold-tolerance traits (CTmin, CCR, SCP) differ by Bartonella infection status

Goal: To test whether Bartonella infection status (B− vs. B+) affects cold tolerance (CTmin, CCR, SCP) in adult Dermacentor variabilis collected in Nova Scotia, And to estimate The Prevalence of *Bartonella* infection in this tick population.

Methods:

In 2025, adult D. variabilis were collected in Nova Scotia through flag sampling and passive submissions. Ticks were identified to species and sex, assigned unique identification codes, and maintained under controlled conditions before experimental testing.



Genomic DNA was extracted from each tick, and Bartonella spp. infection status was assessed via PCR and agarose gel electrophoresis. Samples with a band at the expected amplicon size were classified as Bartonella-positive (B+) samples lacking a band were classified as Bartonella-negative (B−).

Figure 5: Left: Gel documentation system used to visualize fluorescently stained DNA in agarose gels. **Right**: Representative agarose gel image with sample lanes and molecular weight markers (DNA ladder) discrete bands indicate successful amplification and detection in the corresponding samples.

The Cold-tolerance assays:

- CTmin: Ticks were cooled at a constant rate in a programmable cooling bath, and CTmin was recorded as the temperature where coordinated movement was lost (chill coma).
- CCR: Following cold exposure, ticks were returned to room temperature, and CCR (min) was measured as the time to sustained coordinated movement.
- SCP: Ticks were mounted to thermocouples and cooled until an exotherm spike signalled freezing; that temperature was recorded as SCP.

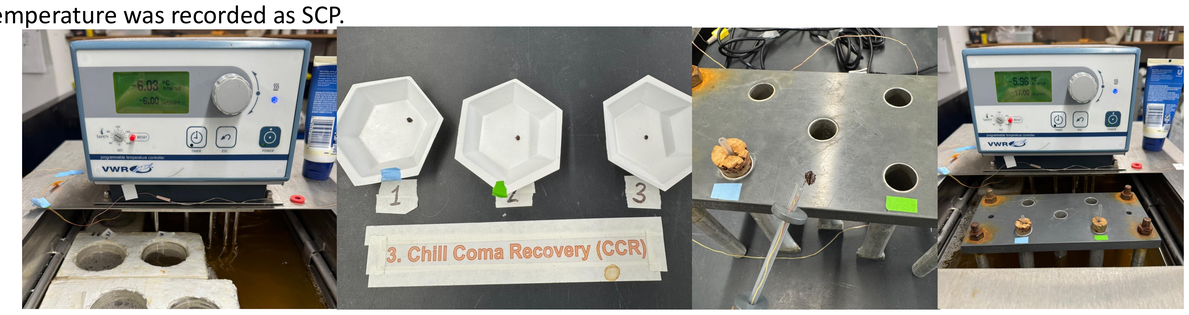
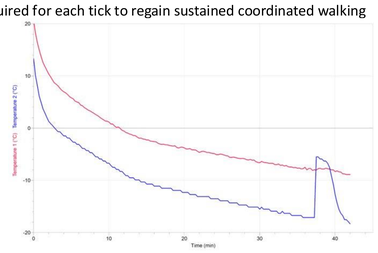


Figure 6: (1) VWR programmable cooling bath used to expose ticks to controlled low temperatures prior to recovery measurements. (2) Individually labelled dishes used to hold and track specimens during CCR trials example IDs 1–3 shown. (3) Pre-cold observation surface where ticks were attached and monitored during SCP. (4) Example CCR run showing the cooling bath and sample rack, with multiple specimens assayed in parallel. CCR was defined as the time required for each tick to regain sustained coordinated walking following cold exposure and return to room temperature.

Supercooling point (SCP) measurements Body temperature was monitored with a thermocouple probe connected to Logger Pro while individual ticks were cooled in a controlled bath. Logger Pro produced a continuous temperature trace for each specimen. SCP was defined as the minimum temperature immediately preceding the exothermic "spike," which indicates the heat release from the exothermic reaction.



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Analysis

For each cold-tolerance trait (CTmin, CCR, SCP), group means ± standard errors were calculated for B− and B+ ticks. Independent two-sample t-tests were used to compare trait values between infection groups.

Why This Matters:

Cold-hardiness traits help determine whether ticks can survive winter conditions and establish, persist, or expand into new areas.
Dermacentor variabilis is common in Nova Scotia and is a potential vector of Bartonella spp., but the relationship between infection and cold tolerance has not been studied in this region.
If infection shifts cold tolerance, it could affect overwinter survival and, in turn, pathogen risk for people and animals.
By comparing cold-tolerance traits in infected and uninfected ticks, this project provides baseline data to help predict how D. variabilis may respond to future winters and climate change in Atlantic Canada.

Hypothesis:

We hypothesized that Bartonella-positive Dermacentor variabilis would be more cold-tolerant than uninfected ticks. Specifically, we predicted that B⁺ ticks would show lower CTmin and SCP values and shorter CCR than B⁻ ticks, consistent with greater cold hardiness.

Results:

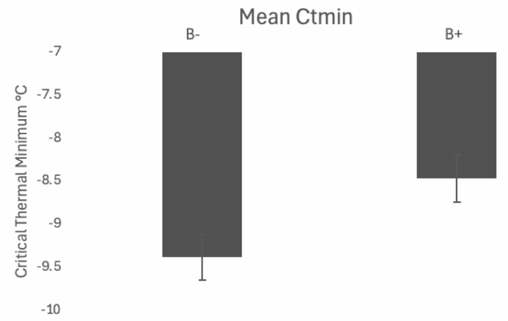

Figure 1. Average critical thermal minimum (CTmin °C) for adult *Dermacentor variabilis*, comparing Bartonella-negative (B−) and Bartonella-positive (B+) ticks.

Table 1: Mean critical thermal minimum (CTmin) for adult *Dermacentor variabilis*, grouped by Bartonella status negative (B−) vs. positive (B+) With data combined across sexes. Values are reported as mean ± standard error (SE) SD indicates standard deviation.

Group	n	Mean Ctmin	SD Ctmin	SE Ctmin
B-	54	-9.378	1.964	0.267
B+	52	-8.463	1.982	0.275

Table 2: Mean chill-coma recovery time (CCR) for adult *Dermacentor variabilis*, grouped by Bartonella status negative (B−) vs. positive (B+) with data combined across sexes. Values are reported as mean ± standard error (SE) SD indicates standard deviation.

Group	n	Mean CCRmin	SD CCRmin	SE CCRmin
B-	54	3.13	5.572	0.758
B+	52	2.68	3.756	0.542

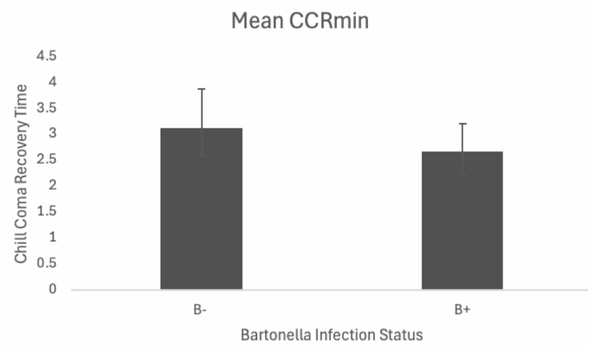

figure 2. Average chill-coma recovery time (CCRT, h) for adult *Dermacentor variabilis*, comparing Bartonella-negative (B−) and Bartonella-positive (B+) ticks.

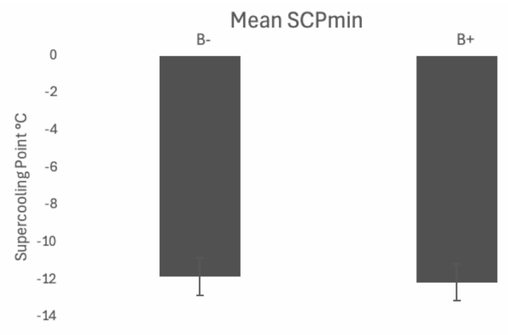

Figure 3. Average supercooling point (SCP, °C) for adult *Dermacentor variabilis*, comparing Bartonella-negative (B−) and Bartonella-positive (B+) ticks.

Table 3: Mean supercooling point (SCP) for adult *Dermacentor variabilis*, grouped by Bartonella status negative (B−) vs. positive (B+) with data combined across sexes. Values are reported as mean ± standard error (SE) SD indicates standard deviation.

Group	n	Mean SCPmin	SD SCPmin	SE SCPmin
B-	54	-11.827	4.179	0.539
B+	52	-12.118	2.904	0.448

Discussion:

Current findings: So far, Bartonella-positive and Bartonella-negative Dermacentor variabilis show only small and inconsistent differences in cold-hardiness traits. Infected ticks tend to have slightly higher warmer CTmin values, while CCR and SCP show the opposite pattern. However, these differences are modest compared to the variation within each group and are not clearly significant yet.
Future directions sampling and pathogens Next steps will focus on increasing sample sizes within each sex and life stage, and screening ticks for additional pathogens *Rickettsia spp.* so we can test co-infection patterns alongside cold-tolerance traits.
Future directions linking traits to ecology Further work will measure body condition or energy reserves and use field overwintering experiments to connect lab-based CTmin, CCR, and SCP to real winter survival. We also plan to compare *D. variabilis* with other tick species in Nova Scotia.

References:

El Nabbout, A., Ferguson, L. V., Miyashita, A., & Adamo, S. A. (2023). Female ticks (*Ixodes scapularis*) infected with *Borrelia burgdorferi* have increased overwintering survival, with implications for tick population growth. *Insect Science, 30*(6), 1798–1809.

El Nabbout, A., Taylor, B. J., Kho, J., Mitton, M., & Rossolimo, T. (2018). The correlation of *Francisella tularensis* on size supercooling of *Dermacentor variabilis*. *International Journal of Biology, 10*(2), 15–22.

Rosendale, Andrew J., et al. "Metabolomic and Transcriptomic Responses of Ticks during Recovery from Cold Shock Reveal Mechanisms of Survival." *Journal of Experimental Biology*, vol. 225, no. 8, Apr. 2022, jeb236497. The Company of Biologists,

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