

Tick Testing and Submission Instructions to NJ PHEL

Changes in this Version

- PHEL is requesting that submitters submit ticks no less than **monthly** during active collections.
- Break up submissions into groups based on tick **species**
- One bag, one species, one form:
 - o Each submission should contain no more than 94 samples of ticks of the same species.
 - o This submission should be contained in one bag, with one submission form inserted into the front pocket. The bag should be labeled with the submission number.
 - o Do not create separate submission forms based on different collections or life stages.

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Tick Testing at PHEL

PHEL will accept and test for pathogens routine submissions of ticks from county agencies enrolled in NJ’s tick surveillance program. PHEL will also coordinate specimen submission should the ticks be sent to the CDC for testing. Pathogen testing in host-seeking, unfed (flat) ticks is recommended for identifying the presence and prevalence of pathogens. PHEL will not test engorged ticks¹ or larvae.

Please contact CDSVectorTeam@doh.nj.gov and james.occid@doh.nj.gov prior to sending tick samples to NJ PHEL.

Tick Testing Overview

PHEL will accept the following species of ticks for testing, and will routinely test for a panel of pathogens by species.

- ***Ixodes scapularis***: tested for *Babesia microti*, *Anaplasma phagocytophilum*, *Borrelia burgdorferi*, *Borrelia miyamotoi*, Powassan virus
- ***Amblyomma americanum***: tested for *Ehrlichia chafeensis*, *Ehrlichia ewingii*, *Rickettsia rickettsii*, Heartland virus, Bourbon virus
- ***Dermacentor variabilis***: tested for *Rickettsia rickettsia*

¹ Partially engorged Asian longhorn ticks found in the environment may be submitted with pre-approval. Contact CDSVectorTeam@doh.nj.gov and james.occid@doh.nj.gov to submit if any are collected

- *Amblyomma maculatum*: tested for *Rickettsia parkeri*
- *Haemaphysalis longicornis*²: tested for *Rickettsia rickettsii*, Heartland virus, Bourbon virus

Number of Ticks to Submit

- For 2023, there are eight counties participating in the NJDOH Tick Surveillance program. The maximum number of ticks each county can submit annually is 1,250. Participating counties may submit **up to 1,250 samples**, regardless of tick species. **Table 1** outlines suggested number of ticks to send for testing per site and life stage based on the number of sites surveyed. **Larval ticks and blood-fed ticks will not be tested for pathogens.**
- **Table 1.** Recommended number of ticks to send for testing as a function of site counts in a county.

Sites	Max Ticks Per Site	Max Adult Ticks	Max Nymphal Ticks
5	250	125	125
6	208	104	104
7	178	89	89
8	156	78	78
9	138	69	69
10	125	62	62
11	113	56	56
12	104	52	52

- Counties are encouraged to collect at least 25 adult and at least 25 nymphal host-seeking *I. scapularis* ticks (or other target tick species determined by the county) per site to estimate pathogen prevalence, irrespective of environmental collection method (ticks collected from hosts are excluded and will not be tested). Increasing the sample size will provide higher confidence on the pathogen prevalence rates for each site.
- **Note:** Typically, 50 ticks per site gives reasonable confidence limits of pathogen prevalence. Regardless of tick species, at least 25 ticks of a given life stage must be tested in a county to display pathogen prevalence rates for that county in CDC maps. Less than 25 ticks can establish pathogen presence, but not prevalence.

² *Haemaphysalis longicornis*: Although this species is currently thought to be competent for *R. rickettsii* and Heartland Virus and other pathogens have been detected in field collections, at this time, *H. longicornis* is not considered to represent a significant public health concern. Counties may submit a subset of *H. longicornis* ticks for pathogen testing, but the focus of the NJ Tick Surveillance Program remains on ticks of significant public health importance.

Pathogen Testing

Submission Instructions

Ticks should be submitted to PHEL on a **bi-weekly or monthly** basis throughout the collection season following the guidelines below. DO NOT batch samples to be submitted any less frequently than once per month.

1. Ticks collected in the field should be sorted and stored by collection site, date, species, and life stage.
2. Ticks **MUST** be surface-sterilized by submersion in 70-95% ethanol before submission- either in the field or before/during sorting.
 - a. Note: It is recommended to collect ticks directly into vials containing 70-95% ethanol.
 - b. Drain ethanol after sterilizing- do NOT submit ticks in vials containing ethanol.
 - c. Identify ticks by species and life stage/sex (**Appendix H of the NJ Tick Surveillance Guide**).
3. Enter data into JerseySurv (**Appendix K of the NJ Tick Surveillance Guide**).
4. Create pools in JerseySurv.
 - a. Pool samples according to species and life stage as below:

Species	# of adults/pool	# of nymphs/pool
<i>I. scapularis</i> <i>A. maculatum</i>	1	1
<i>A. americanum</i>	1	up to 5
<i>H. longicornis</i> <i>D. variabilis</i>	up to 5	up to 10

5. Create submission in JerseySurv
 - a. Break up submissions into groups based on tick **species**, with no more than 94 samples per submission.
 - b. Select only one test request for all samples in one submission form, based on the tick species:

Tick Species	Panel to Select in JerseySurv
<i>I. scapularis</i>	T-IS
<i>A. americanum</i> <i>H. longicornis</i>	T-AA
<i>D. variabilis</i>	T-RR
<i>H. longicornis</i>	T-HL
<i>A. maculatum</i>	T-AM

- c. One bag, one species, one form:
 - i. Each submission should contain no more than 94 samples of ticks of the same species.
 - ii. This submission should be contained in one bag, with one submission form inserted into the front pocket. The bag should be labeled with the submission number.
 - iii. Do not create separate submission forms based on different collections or life stages.
 - d. If you are submitting multiple species for testing, create multiple submission forms:
 - i. For example, if you have 24 pools of *A. americanum* ticks (regardless of life stage or collection location) they can all be submitted in one submission.

- ii. If you have 24 pools of *A. americanum* and 24 pools of *I. scapularis*, create two submissions.
 - e. If you have a large submission, with more than 94 ticks of the same species, create two separate submissions:
 - i. For example, if you have 100 *I. scapularis* ticks, create one submission with pools #1-94, and a second submission with pools #95-100. This facilitates processing at the lab.
 - ii. If you are collecting large volumes of ticks, we would encourage you to inquire about increasing the frequency of your submissions instead of batching them to send once a month.
 6. Place sorted ticks to be submitted for testing in **green vials provided by PHEL** containing one copper BB.
 - a. Place only one tick per vial (unless pooling, see above)
 - b. Label the vials using the pool ID number generated by JerseySurv for each submission.
 - c. Place the label on the side of the tube (wrapped horizontally) and write the pool ID number on top of the vial (same as mosquito submissions)
 - d. Place labeled vials into biohazard sample bags and insert the submission form into the bag.
 7. Ticks can be shipped at room temperature. Preferably, ticks should be stored frozen and shipped on ice packs (using state courier service or individual deliveries).
 - a. **Note: Since tick should have been submerged in alcohol prior to submission (step 2), there is no need to add alcohol to the submission tube.**

CDC Submission Criteria

If counties are collecting a high volume of *I. scapularis* ticks such that submissions to PHEL will exceed the 1,250 maximum number of ticks per season, please contact NJ PHEL for guidance. Some submissions may qualify for direct submission to CDC, and we would request that you submit ticks following the most up to date CDC instructions and select a different testing panel (TC-I) for the pools.

Contact CDSVectorTeam@doh.nj.gov, James.Occi@doh.nj.gov and Dana.Woell@doh.nj.gov for approval prior to submitting ticks for direct submission to CDC.