TICK SURVEILLANCE RECOMMENDATIONS

Version 4.0

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BACKGROUND

Tick surveillance is intended to monitor changes in the distribution and abundance of ticks and the presence and prevalence of tickborne pathogens to provide actionable, evidence-based information to clinicians, the public, and public health policymakers. Key questions address when and where humans are at risk for exposure to ticks and tickborne pathogens.

New Jersey (NJ) is in the top 20% of states reporting the largest numbers of tickborne disease cases, with 21,284 cases reported between 2022 and 2024. Tickborne diseases reported in NJ include anaplasmosis, babesiosis, *Borrelia miyamotoi*, ehrlichiosis, Lyme disease, spotted fever group rickettsiosis, Powassan virus, and tularemia. Additionally, there have been 1,039 reported cases of Alpha-Gal Syndrome during the same timeframe.

The two tick species of greatest public health importance in NJ are *Ixodes scapularis* (blacklegged or deer tick) and *Amblyomma americanum* (lone star tick). In 2022, the first established population of *Amblyomma maculatum* (gulf coast tick) was identified in NJ and since then, ticks have been reported in multiple counties. *A. maculatum* is a vector of *R. parkeri*, which is one of the spotted fever rickettsioses, but the burden of these infections in NJ is unknown. *Dermacentor variabilis* (American dog tick) is a vector of *R. rickettsii*, the causative agent of Rocky Mountain Spotted Fever elsewhere in the US, but tick testing in NJ has failed to detect human tickborne disease pathogens. *Haemaphysalis longicornis* (longhorned tick) is present throughout the state, although it is not currently considered a human health concern.

The discovery of the longhorned tick in Hunterdon County in 2017 revealed the need for tick surveillance in New Jersey. A species native to East Asia, the Asian longhorned tick is a serious livestock pest associated with several human tickborne diseases in other parts of the world, including anaplasmosis, ehrlichiosis, and severe fever with thrombocytopenia syndrome. Although the Asian longhorned tick was identified in November 2017, subsequent findings show that the Asian longhorned tick was present in New Jersey as early as 2013. Thus, the arrival of the Asian longhorned tick is an example of how statewide tick surveillance is necessary to protect human and animal health.

NJDOH has limited funds to support a small number of counties to establish or maintain routine tick surveillance activities to provide statewide surveillance data. Additionally, counties that conduct active tick surveillance using local resources can contact NJDOH and request pathogen testing. NJDOH/NJDEP will supplement surveillance conducted by county programs as resources allow to implement statewide priority surveillance strategies. Data collected from tick surveillance activities can identify areas where ticks of public health importance are located in NJ and establish tick abundance and infection rates. NJDOH, through the Public Health and Environmental Laboratories (PHEL), will provide tick testing for county agencies participating in the Tick Surveillance Program.

SURVEILLANCE OBJECTIVES

The surveillance goals of the NJDOH Tick Surveillance Program include:

1. Classifying county status for ticks of public health concern: reported, established, or no data available
2. Classifying county status for the presence of specific pathogens in ticks: present or no data available
3. Generating estimates for the local prevalence of specific pathogens by species
4. Generate estimates of local density of host-seeking (infected) nymphs or adults
5. Conducting targeted surveillance to address priority issues

SITE SELECTION

Agencies that haven't conducted tick surveillance previously should use existing data sources (e.g., JerseySurv) and/or sample a broad range of sites across the county to detect the presence or absence of medically important tick species. While there is no set number of sites counties should sample (recommendations are provided in the following paragraph), it is encouraged to sample broadly across various habitats based on which ticks are being sought. However, sites should be selected based on their proximity to population centers, rather than a remote location in the county. Priority should be placed on municipalities where tick-borne diseases are most prevalent or demonstrate increasing incidence. Agencies conducting tick surveillance are encouraged to coordinate with local public health agencies to identify priority areas within a county.

Counties participating in the NJDOH Tick Surveillance Program should focus efforts on at least one of the ticks of greatest public health importance in New Jersey (*I. scapularis*, *A. americanum*) based on local needs. While *D. variabilis* is a tick of medical importance in the US, NJDOH has not identified any human pathogens in ticks collected through the state program and as a result, they will not be prioritized for testing at PHEL starting in 2025.

There are two approaches to the collections:

1. A minimum of two collection sites per year should be sampled using standardized density sampling. In addition to these two sites, 5-10 additional exploratory sites should be visited **during the nymphal and adult peaks**, with the aim of identifying the presence of ticks and pathogen prevalence.
2. **Alternatively**, counties can collect on at least five sites using the standardized density sampling approach. Counties using this approach are not required, but are encouraged, to do exploratory sampling.

Sites, whether exploratory or not, should be visited **during the nymphal and adult peaks**, with the aim of identifying the presence of ticks and pathogen prevalence. The greater the geographic areas sampled, the more representative the data will be.

After density estimates have been generated for nymphal and adult tick populations in one site, counties should consider sampling the same site over at least three years. After three years, counties can continue monitoring at existing sites or focus on performing density sampling in other sites. Sites with tick density estimates already established may be revisited in rotating 3–4 year intervals.

Considerations for site selection may be based on several criteria:

1. Prevalence of habitat-specific to tick targeted for surveillance (**Appendix A**).

2. Evidence of ticks based on prior sampling.
3. Location in the county, to select sites representing different parts of the county when possible. Select sites that have a high probability for long-term use, preferably on public property.

Note: Before selecting a site, permission should be obtained from appropriate parties. Contact information should be kept on file.

Size of area to sample

The density of host-seeking nymphal or adult ticks varies spatially and temporally. Therefore, for sites where density sampling will be conducted ensure that the sampling area in one site is expansive (spanning at least 750 m^2 of linear transects, a 750 m^2 grid, or 50 transects of 15 m). This will then allow for a more representative sample of the density of host-seeking (infected) nymphs or adults. The distance sampled can be assessed using one of two methods:

1. Setting fixed sampling grids where flags, stakes, or other objects are used to mark the start and endpoints of each measured length of the transect, or
2. Use a measuring wheel with meter as the unit of measurement.

For exploratory sites (sites where density sampling is not performed), the sample area can vary. It is recommended to create 100-500m (recommended: 350m) transects per sampling sites, though ideally sampling 750 m^2 per exploratory site can ensure that site is sufficiently sampled. These exploratory sites should be visited at least twice: one time each during the nymphal and adult peaks. Counties should aim to collect as many ticks as possible at each site, with the goal of collecting 25-50 ticks (nymph or adult) in order to generate presence/absence data as well as pathogen prevalence rates.

When to sample

Counties participating in the NJDOH Tick Surveillance Program should sample between the nymphal and adult peak times for the ticks of interest (**Appendix A and Figure 1-2**). For standard tick surveillance sites, sampling should be conducted at least twice during each nymphal and adult peak. For agencies not receiving funds from NJDOH for sampling, nymphal tick collections should be prioritized between late May and early July.

Summary of seasonal peaks by species and life stage

Tick	Seasonal Peak
***I. scapularis* nymph**	Late May through early July
***A. americanum* nymph**	Late May through early July
***I. scapularis* adults**	Late October through December
***A. americanum* adults**	Mid-May through late June
***D. variabilis* adults**	Early May through mid-June
***A. maculatum* adults**	June – September (limited data)
***A. maculatum* nymphs**	Unknown

Collections should be conducted during favorable weather, with air temperature >40°F and wind speed <20 miles/hr. Sampling should not be conducted when it is raining, when the vegetation is wet enough to saturate the tick flag, or when the weather is unseasonably cold or windy. Scheduling collections at different times of day at each site can reduce temporal effects during flagging. As weather conditions, time, and resources allow, sample each site every two weeks during the months when nymphal or adult ticks are active.

FIELDWORK

Attire

Guidance from: Centers for Disease Control and Prevention (CDC). (2020). Guide to the surveillance of metastriate ticks (Acari: Ixodidae) and their pathogens in the United States. *Division of Vector-Borne Diseases, CDC, Atlanta, GA and Ft. Collins, CO. https://www.cdc.gov/ticks/pdfs/Tick_ surveillance-P.pdf*, 19-20.

Individuals involved in collections should wear white or light-colored clothing or cotton coveralls, sturdy, tall, laced shoes (like hiking boots), and tall white socks pulled over the bottom of the pants, securing them with tape applied at the sock-pant junction. Tyvek coveralls should be avoided, especially in hot weather, due to the chances of overheating and dehydration. Tape can also be used to cover shoe eyelets. In addition, an EPA-registered insect repellent that contains DEET, picaridin, R3535, Oil of Lemon Eucalyptus (OLE), para-menthane-diol (PMD), or 2-undecanone may be used. Clothing and boots should be treated with 0.5% permethrin (ensure to apply permethrin away from the tick collection equipment).

When collecting ticks, do not let the drag/flag/sweep touch the treated clothing. Additionally, do not touch treated clothing with hands when collecting and keep forceps/vials out of pockets. Forceps, vials, markers, and other miscellaneous equipment can be stored in a belly pack. Finally, if conducting tick surveillance in an area where public hunting is allowed, it is recommended that surveillance workers wear bright (recommended color: hunter orange) safety vests and caps.

Supplies

Refer to **Appendix B** for a list of recommended supplies needed for field collections and **Appendix C** and **D** for materials and instructions for constructing a tick drag or flag.

TICK COLLECTION

While there are several tick collection methods, not all are appropriate for estimating tick density calculations (**Table 1**). As a result, counties participating in the NJDOH Tick Surveillance Program should use flagging or dragging techniques when conducting density sampling. Tick dragging is the recommended sampling approach, but local programs can determine the optimal method based on the habitat in their sampling sites. Regardless of the sampling approach, counties should be consistent.

- **Tick Dragging:** Tick dragging is a tick collection method whereby a piece of cloth, typically with an area of $1m^2$ is attached to a dowel, with a rope affixed to each end of the dowel, is pulled along behind the collector.
- **Tick Flagging:** Tick flagging is a tick collection method where a piece of cloth, typically with an area of $1m^2$ is attached to a long pole in the manner of a banner and swept in front of or to the side of the operator.

- **Tick Sweeping:** Tick sweeping is like tick flagging but uses a smaller piece of cloth, such as 0.25m^2, that is swept in front of or to the side of the operator.

Table 1. Comparison of tick collection methods

Collection Method	Objective: Classify county status	Objective: Presence/Prevalence of pathogens in ticks	Objective: DON/DIN or DOF/DIF	Objective: Phenology
Dragging/Flagging/Sweep	Acceptable	Acceptable	Acceptable	Acceptable
Walking	Acceptable	Acceptable	Not Acceptable	Acceptable
CO_2 traps	Acceptable	Acceptable for presence, but not prevalence	Not Acceptable	Not Acceptable
Ticks collected from deer	Acceptable	Acceptable for presence, but not prevalence	Not Acceptable	Not Acceptable
Ticks collected from small- or medium-sized mammals, birds, lizards	Acceptable	Acceptable for presence, but not prevalence	Not Acceptable	Acceptable
Ticks from people/pets	Acceptable, if travel history is accounted for	Acceptable for presence, but not prevalence	Not Acceptable	Not Acceptable

Source: Centers for Disease Control and Prevention (CDC). (2020). Guide to the surveillance of metastriate ticks (Acari: Ixodidae) and their pathogens in the United States. *Division of Vector-Borne Diseases, CDC, Atlanta, GA and Ft. Collins, CO. https://www. cdc. gov/ticks/pdfs/Tick_ surveillance-P. pdf,* 7.

Tick Collection Method and Data Recording

When in the field, the Field Collection Form (**Appendix G**) can be used to ensure all the required data are collected and reduce recall bias. All collections must ultimately be recorded in the JerseySurv Tick Module. Field collectors should record start and end measurements for time, temperature, wind speed, and humidity when possible. While flagging/dragging for nymphs, the drag/flag must have maximum contact with the ground and leaf litter. Depending upon the weather (air temperature, wind speed, humidity), adult ticks may be found anywhere from the level of the leaf litter up to one meter high on vegetation. Therefore, while sampling for adults, vegetation up to approximately one meter in height should be sampled, but low-lying vegetation should not be avoided purposefully. For collectors using tick drags, the drag can be picked up from the dowel to sample over the vegetation, ensuring that the drag goes over the entire perimeter of the vegetation and the ground near it. Weights (e.g., metal washers or chains) may be sewn into the trailing edge of the tick drag/flag to increase contact between the fabric and vegetation. In cases where vegetation is higher than one meter, or if the arrangement of the vegetation (e.g., shrubbery hedges) makes sampling difficult, then the collector can sample around the vegetation. Modified handles (e.g., dowel or rope) may be used to increase maneuverability. The tick drag/flag should be moved across vegetation or leaf litter.

Even if many ticks are obtained from an area, do not repeatedly sample that same piece of ground that day. Sampling multiple times on the same piece of ground will skew the tick density data. However, several simultaneous drags/flags in different areas of the same site (i.e. different sections of a single park) can ensure the reliability of the tick collection data.

Because ticks can easily drop off the drag/flag, inspecting the cloth at regular 10-15m intervals is important . Drags/Flags should be checked systematically, and all parts of the cloth should be examined, including the

leading edge, ropes, and seams. Samplers should also inspect their hands at each cloth check and include any ticks found.

NOTE: When conducting tick collection, ideally there should be multiple collectors per trip, so that they may check one another's clothing for ticks.

Area to Sample per Sampling Session

In each standardized density sampling session, a minimum of 750 m^2 should be dragged/flagged. There are several different methods recommended to measure the area being sampled:

1. Before collecting, to keep track of the area sampled, measure the distance, marking 10-15 m lengths with stake flags. When in the field, check the drag/flag every 10-15 m, keeping track of the number of times the drag/flag was checked using a tally counter. Record this number next to each collector's name on the field collection datasheet.
2. Before collecting, measure out 10 m in length. Walk the measured length 4 times, counting your steps each time. Calculate your step count by taking the average number of steps for the 4 walks. When in the field, check the drag each time you walk your average step count, keeping track of the number of times the drag/flag was checked using a tally counter. Record this number next to each collector's name on the field collection datasheet.
3. Use a measuring wheel with meter as the unit of measurement. When in the field, check the drag/flag every 10-15 m as per the measuring wheel. After dragging, record the final measurement to each collector's name on the field collection datasheet.

The total area sampled can be calculated as follows: # of times the drag/flag was checked x the distance traveled by each collector before checking their drags/flags (10-15 m lengths).

Handling Field Collected Ticks

Ticks can be removed from the drag/flag using fine-point forceps and placed in a vial with 70-95% ethanol. Alternatively, ticks could be placed in a zipper-top plastic bag. Using tape to remove the ticks is not recommended unless larvae or nymphs are too numerous on the drag. If ticks are removed using tape, collectors can place the tape in a zipper-top plastic bag and remove the ticks after field collection. Make sure to label the bag or vial with the site name, transect number, collection method, county, and date.

SUPPLEMENTAL TICK COLLECTION METHODS

Dragging/flagging are preferred methods for tick surveillance where tick species are known to be present, and the focus is on monitoring prevalence density. However, CO_2 trapping and sweeping are useful techniques to supplement surveillance efforts when used to establish the presence of tick species in new areas.

Carbon Dioxide (CO_2) Trapping

Carbon dioxide traps work on the premise that ticks have well-developed chemoreceptors and are attracted to carbon dioxide to find a host. Traps consist of a solid base to hold dry ice (a solid form of carbon dioxide) within an insulating material surrounded by a sticky tape to capture ticks attracted to the carbon dioxide released as the dry ice sublimates. The trap was developed originally to collect lone star ticks (*Amblyomma americanum*), which display a more aggressive and mobile host-seeking behavior. Asian longhorned ticks are also attracted to CO_2. While CO_2 traps capture *I. scapularis*, they are less effective than drag sampling or flagging. Because of its inefficiency at collecting *I. scapularis*, this trapping method is not recommended for targeting this species. However, this sampling method provides good spatial precision for documenting other tick species' occurrence or presence. Additionally, ticks collected using CO_2 traps can be used to detect the presence of pathogens.

Tick Collection Method

CO_2 traps work best on windless days and early in the morning or later in the day when there is less wind. Place the trap flat among vegetation, not in the open sun. Place white fiberboard or plywood underneath (3-4 inches surrounding trap) covered in double-sided sticky tape to collect ticks. Traps should be set at least 2 hours and up to 24 hours before retrieval.

After at least 2 hours (and up to 24 hours), check traps, remove ticks from tape with forceps, and place them in a vial with 70-95% ethanol or alternately, in a plastic zipper-top bag. If removal in the field is not possible, seal the tape containing ticks in the plastic bag. Label the bag with the site name, collection method, county, and date (do not mix ticks collected by different collection methods).

Tick Sweeping

Sweeping is a method similar to tick flagging. While mainly used to establish the presence/absence of ticks in a particular area, they can also be used for tick density estimates. The technique is particularly helpful when the presence of a tick in an area has not been documented before. Therefore, tick sweeping is useful during exploratory data collection, especially in determining whether an area is appropriate for the surveillance of a particular tick species. When using tick sweeps, collectors must keep in mind the area of the cloth and ensure the sampling area is collected. While tick sweeps can be used for tick density estimates, it is still strongly recommended that collectors use tick drags or tick flags for standardized density sampling. Instructions on how to create a tick sweep are included in **Appendix E**.

When entering collection information in JerseySurv, please use either FLAG or DRAG as the method in JerseySurv if performing tick density sampling so it gets reported to the CDC properly. The decision to use DRAG or FLAG will depend on how the tick sweep is used. If simply dragged along vegetation, use DRAG. If collecting from the side or in front of the collector, use FLAG.

Site Selection

Different tick species are associated with different habitats. Therefore, counties should find an appropriate site as outlined in **Appendix A**.

Size of Area to Sample

Because tick sweeping is primarily used for exploratory data collection, transects established are typically smaller than what is recommended by the CDC. For each county, potential collection sites should consider 100m-500m transects per site, when possible.

Tick Collection Method

Each transect will be sampled with a tick sweep held to the side of the collector at a slow, steady pace. The tick sweep should be checked every 10-15m for ticks. Additionally, the collector should also perform a tick check on themselves. Ticks can be collected by using fine-tipped forceps and then placed in a microcentrifuge vial filled with ethanol (or zipper-top bag) with a label containing the date/time of the start/end of the collection, collector's name, county, site number, and transect number. If collecting using tape, the tape can be placed in the zipper-top bag, and ticks can be removed from the tape after field collection.

TICK IDENTIFICATION AND REPORTING

After field collection, ticks should be sorted and identified to species and life stage using published taxonomic keys (see **Appendix H**). Enter total collection data (abundance data) in the Field Data Collection Form (**Appendix G**) or directly into JerseySurv under "Tick-Abundance". After identification, ticks that will be submitted for testing should be placed in vials (*without ethanol* – see Tick Testing instructions) according to collection site, date, species, and life stage. All collections and pool data must be entered into JerseySurv (instructions in **Appendix K**).

If an agency is unable to speciate (or cannot identify to genus and life stage), they can place samples in a vial with 80% ethanol with a label "<GENUS NAME> – unidentified" (e.g., "Dermacentor – spp") and send to PHEL with the following information: collector, date of collection, and location of collection (site name and GPS coordinates). After PHEL has identified the species, the county program will be responsible for updating species information in JerseySurv.

If an agency suspects they have identified a new tick species, not endemic to New Jersey, they should send the specimen to PHEL for confirmatory identification and notify the CDS Vector Team at **cdsvectorteam@doh.nj.gov**. Instructions can be found on **Appendix I**.

TICK TESTING

Ticks are tested at PHEL and if indicated, PHEL will coordinate additional testing at CDC. Pathogen testing in host-seeking, unfed (flat) ticks is recommended for identifying the presence and prevalence of pathogens. Results from pathogen testing in fed ticks should be considered with caution because: 1) in some cases, ticks can acquire pathogens from hosts while feeding and become infected, but not be able to maintain infection through the molt to the next life stage, and 2) infection rates derived from blood-fed ticks or from hosts is not representative of infection rates in host-seeking ticks.

Tick Testing Overview

PHEL tests nymphs and adult ticks for human pathogens based on tick species as described below. In 2025, agencies should submit nymphs and adult FEMALE ticks for pathogen testing.

- ***I. scapularis***: *Babesia microti, Anaplasma phagocytophilum, Borrelia burgdorferi, Borrelia miyomotoi,* Powassan virus
- ***A. americanum***: *Ehrlichia chafeensis, Ehrlichia ewingii, Rickettsia rickettsii,* Heartland virus, Bourbon virus
- ***A. maculatum***: *Rickettsia parkeri*
- ***H. longicornis***: *Rickettsia rickettsii, Heartland virus, Bourbon virus*
- ***D. variabilis***: *Rickettsia rickettsii (***note: for the 2025 tick collection season, *D. variabilis* ticks will not be routinely tested. Agencies should consult with CDS to discuss testing needs***)*

Number of Ticks to Submit

For 2025, there are eight counties participating in the NJDOH Tick Surveillance program. The maximum number of tick pools each county can submit annually is 1,000[1]. For example, if ticks are collected at 5 sites, aim for no more than 200 ticks/site (100 adult/100 nymph). **Larval ticks and blood-fed ticks should not be submitted for testing; for 2025, when submitting adult ticks, agencies should send only female ticks.**

Agencies are strongly encouraged to collect at least 25 female and 25 nymphal host-seeking *I. scapularis* ticks (or other target tick species/life stage as determined by the agency) per county per calendar year 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.

Submitting Ticks for Testing

Agencies new to the NJDOH Tick Surveillance Program should contact CDSVectorTeam@doh.nj.gov to discuss tick testing needs prior to sending tick samples to PHEL.

Timing

Tick submissions to PHEL should start in May each year. Earlier collections (January-April) should be stored frozen (see below). Ticks should be submitted to PHEL on AT LEAST a monthly basis throughout the collection seasons (nymphal, adult). For large volume submissions (>94 pools), agencies are encouraged to send more frequent submissions. At the end of the collection season(s), agencies should notify CDSVectorTeam@doh.nj.gov when all submissions have been sent to PHEL and for all annual collections, no later than the end of the calendar year.

Specimen submission instructions

1. Ticks **MUST** be surface-sterilized by submersion in 70-95% ethanol before submission- either in the field or before/during sorting.

1 *D. variabilis* should not be submitted for testing in 2025. *H. 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 (not to exceed 250/year), as the focus of the NJ Tick Surveillance Program remains on ticks of significant public health importance.

a. Note: It is recommended to place ticks collected in the field directly into vials containing 70-95% ethanol.

2. Drain ethanol after sterilizing- do NOT submit ticks in vials containing ethanol.
3. Once total collection data is entered into JerseySurv (see Tick Identification and Reporting), pool samples by species and life stage:

Species	# of adults/pool	# of nymphs/pool
I. scapularis ***A. maculatum***	1	1
A. americanum	1	up to 5
H. longicornis ***D. variabilis* (no routine testing in 2025; consult with CDS)**	Up to 5	10

4. Enter submissions in JerseySurv (**Appendix K**)
    a. **<u>One bag, one species, one form</u>**:
        i. Each submission should contain no more than 94 ticks of the same species.
        ii. Each submission should be contained in one bag, with one submission form inserted into the front pocket and the bag labeled with the submission number.
        iii. Do not create separate submission forms based on different collections or life stages. *Individual pools of nymphs and female ticks of the same species and from various collection sites/dates can be combined in one submission*.

    b. Select only one test panel for each submission, based on the tick species. Do not select "Panel TC-I" without prior approval from PHEL – this is for CDC send-outs.

Tick Species	Test Panel
I. scapularis	T-IS
A. americanum ***H. longicornis***	T-AA
A. maculatum	T-AM
D. variabilis	T-RR (*no routine testing in 2025, consult with CDS*)

    c. To submit multiple species for testing, or to submit >94 ticks of the same species, create multiple submission forms. Submission examples:
        i. For 24 pools of *A. americanum* ticks (regardless of life stage or collection site/date) combine all in one submission.
        ii. For 24 pools of *A. americanum* and 24 pools of *I. scapularis*, create two submissions, one for each species.
        iii. For 100 *I. scapularis* ticks, create one submission with pools #1-94, and a second submission with pools #95-100.

5. Place pooled ticks (see Specimen Submission Instructions #3) in **green vials** **provided by PHEL** containing one copper BB.
    a. Label the vials using the pool ID number generated by JerseySurv for each submission.
    b. Place the label on the side of the tube (wrapped horizontally) and <u>write the pool ID number on the top of the vial</u> (same as mosquito submissions)

c. Place labeled vials into biohazard sample bags and insert the submission form into the bag.

6. Ticks can be shipped to PHEL for testing at room temperature. If shipping is delayed >2 weeks from the date of collection, they should be stored frozen and shipped on ice packs (using state courier service or individual deliveries). Since ticks should have been submerged in alcohol prior to submission (step 1), alcohol should NOT be added to the submission vial.

CDC Submission Criteria for *Ixodes scapularis*

If agencies collect a large volume of *I. scapularis* ticks that would exceed the maximum number of ticks PHEL can test per season, contact NJDOH (CDSVectorTeam@doh.nj.gov**,** James.Occi@doh.nj.gov, Dana.Woell@doh.nj.gov) for guidance. Large submissions (25-50 nymphal or female per site per calendar year) may be eligible for testing at CDC and have modified submission instructions.

REUSING FIELD COLLECTION VIALS

Agencies are encouraged to use vials with ethanol to collect ticks in the field. To reduce plastic waste, counties are encouraged to clean and reuse these vials once ticks have been processed and sent for testing. Marks and labels on a vial can be wiped off using alcohol.

The **green vials** provided by PHEL should NOT be used for collecting ticks in the field. The **green vials** are only for submitting ticks to PHEL for testing.

DATA MANAGEMENT

Agencies should enter all collection data and pools submitted for testing into the Tick Module of the JerseySurv electronic surveillance system, referring to **Appendix K: JerseySurv Tick Module Entry**. PHEL will enter tick test results into JerseySurv. The NJDOH Communicable Disease Service will report surveillance and testing data to CDC via ArboNet.

APPENDICES

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Appendix A: Tick Species by Seasonality and Habitat in New Jersey

Tick	Life Stage	Seasonality	Habitat
Lone star tick (*A. americanum*)	Larvae	Late April – Early May[2]; Early August – Late October[2]	**Generally:** Forests, woodlands, fields, and ecotones. **Larvae:** Leaf litter **Nymphs/Adults:** Tall grass in shade or at the tips of low-lying plants.
	Nymph	Mid-March – Mid-September[1]	
	Adult	Mid-March – Mid-July[1]	
Gulf Coast tick (*A. maculatum*)	Larvae	Unknown	**Generally:** Drier environments, recently excavated, and open fields, grass prairies, coastal banks, dunes, sea cliffs
	Nymph	Unknown	
	Adult	June – September[4]	
American dog tick (*D. variabilis*)	Larvae	Early March – Late September[2]	**Generally:** areas with little or no tree cover (grassy fields, scrubland, walkways, trails), tall grass, ecotones, low-lying brush, and twigs.
	Nymph	Early March – Late September[2]	
	Adult	Mid-February – Mid-August[1]	
Blacklegged tick (*I. scapularis*)	Larvae	Early August – Late September[2]	**Larvae/Nymphs:** Moist leaf litter in wooded areas or forest edges **Adults:** Mostly edge of woods
	Nymph	Early March – Mid September[1]	
	Adult	Early September – Late June[1] (Depending on temperature)	
Asian longhorned tick (*H. longicornis*)	Larvae	Early April – Early June[3]; Early July – Late October[3]	**Generally:** Meadows, paddocks, shrubberies, unmaintained grass, and grassy areas near forests
	Nymph	Early March – Late September[3]	
	Adult	Mid-May – Early September[3]	

[1] Jordan, R. A., & Egizi, A. (2019). The growing importance of lone star ticks in a Lyme disease endemic county: passive tick surveillance in Monmouth County, NJ, 2006–2016. *PLoS One, 14*(2)*,* e0211778.

[2] Occi, J. L., Egizi, A. M., Robbins, R. G., & Fonseca, D. M. (2019). Annotated list of the hard ticks (Acari: Ixodida: Ixodidae) of New Jersey. *Journal of medical entomology*, *56*(3), 589-598.

[3] Bickerton, M., McSorley, K., & Toledo, A. (2021). A life stage-targeted acaricide application approach for the control of Haemaphysalis longicornis. *Ticks and Tick-borne Diseases, 12*(1), 101581.

[4] Estimates based on limited collection data from NJDOH Tick Surveillance Program, 2022-2024.

Note: Some months for seasonality are approximated when ranges between seasons are described from the literature rather than between months.

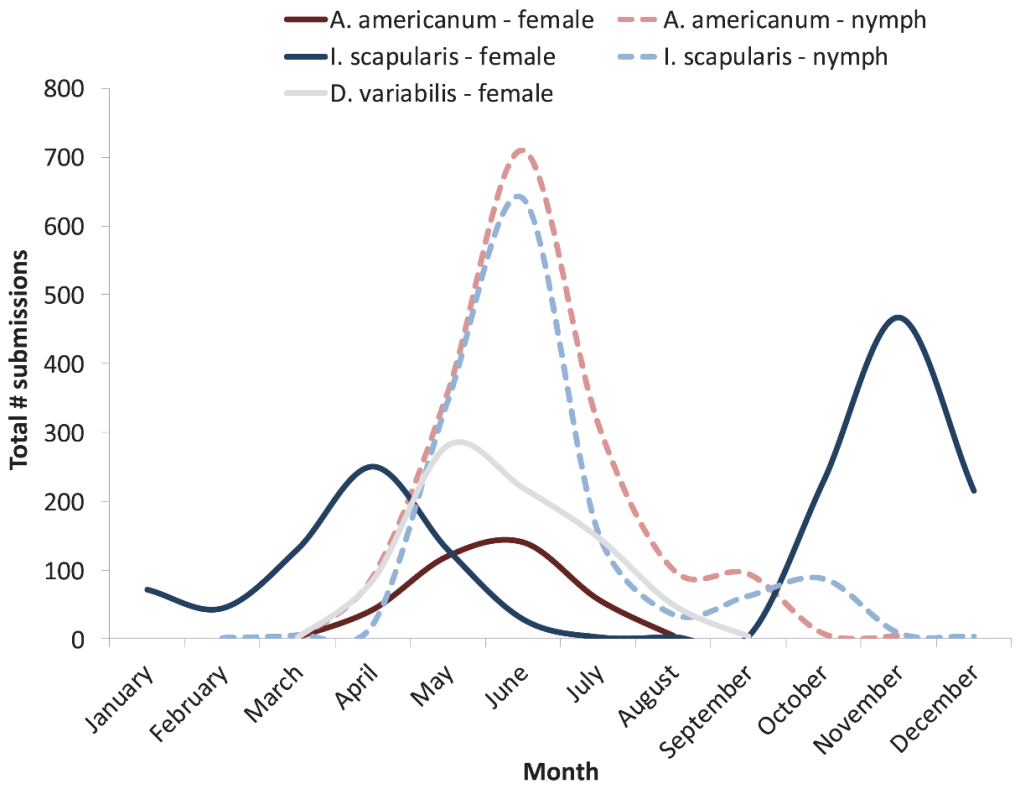


Figure 1. Seasonal phenology of three medically important tick species in New Jersey (Jordan et al., 2019).

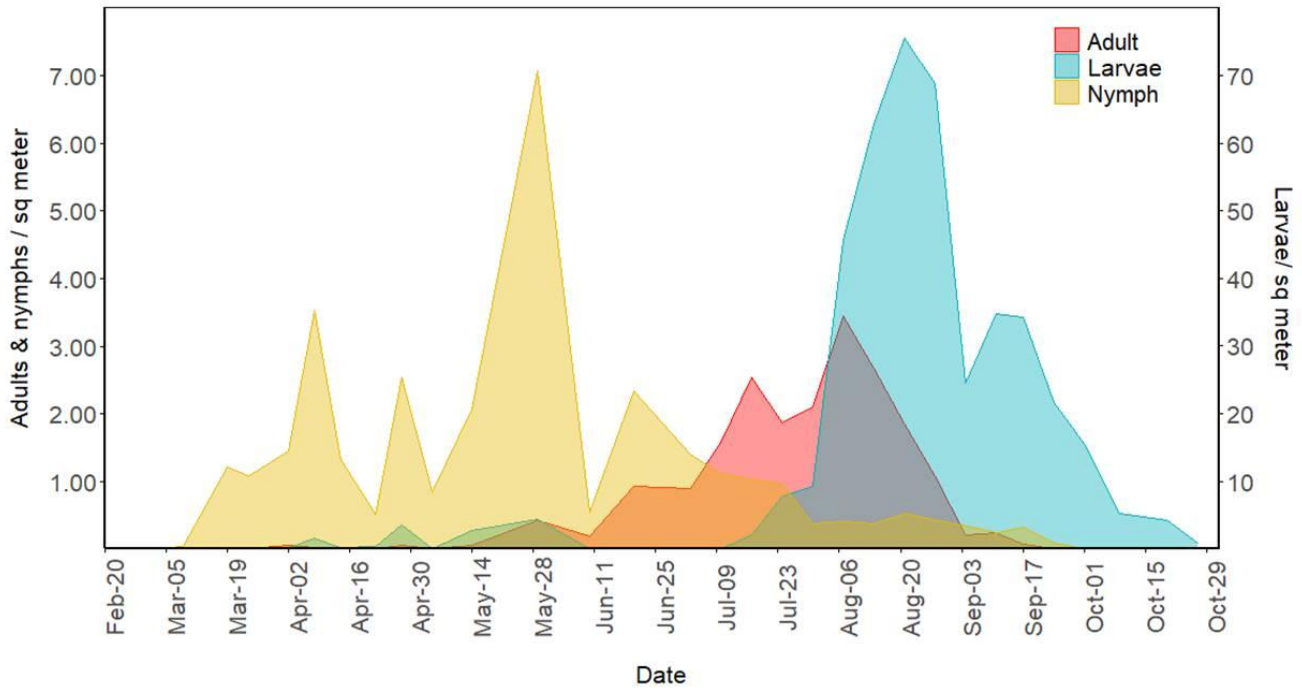


Figure 2. Seasonal phenology of *H. longicornis* (Asian longhorned tick) in New Jersey (Bickerton, 2021).

Appendix B: Supply List

Recommended supplies for tick surveillance and testing:		
Tall white socks	Hiking boots	Long sleeve shirts/pants and/or white cotton coveralls
Permethrin	EPA registered insect repellent effective against ticks	Bright reflective vest or cap
Data collection sheets (See **Appendix G**)	Pen or pencil	Sharpie permanent markers
Kestrel anemometer that measures temp., wind speed, and humidity	Painter's tape	Fine-point forceps
2mL vials for field collections*	2mL vials for tick testing submission* (green vials in 2025)	Tick Drag/Flag (See **Appendix C** and **D**)
70-95% Undenatured ethanol*	Plastic zipper-top bags	Stake flags
**Items may be provided by NJDEP or NJDOH*		

Additional supplies that may be helpful:		
GPS or phone-enabled GPS app	Clipboard	Cell phone
Tally counter	Area map	

Appendix C: Tick Drag Construction

Adapted from: Centers for Disease Control and Prevention (CDC). (2020). Guide to the surveillance of metastriate ticks (Acari: Ixodidae) and their pathogens in the United States. *Division of Vector-Borne Diseases, CDC, Atlanta, GA and Ft. Collins, CO. https://www.cdc.gov/ticks/pdfs/Tick_ surveillance-P. pdf*, 50-53.

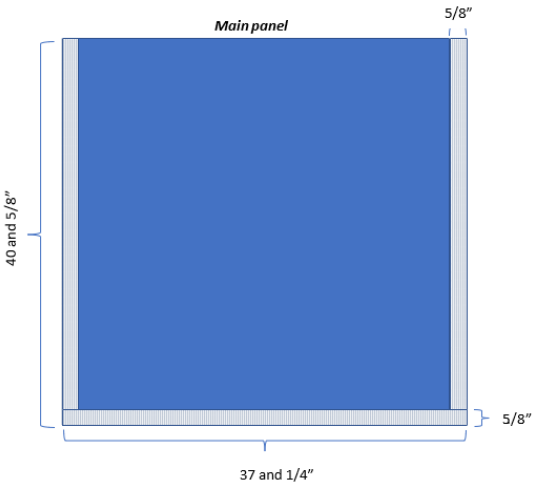
Reference: Newman, B. C., Sutton, W. B., Wang, Y., Schweitzer, C. J., Moncayo, A. C., & Miller, B. T. (2019). A standardized method for the construction of a tick drag/flag sampling approach and evaluation of sampling efficacy. *Experimental and Applied Acarology, 79*(3), 433-446.

Materials for 1 tick drag
Cotton white flannel fabric, corduroy, or cotton bleached white muslin fabric (at least 37 and 1/4" wide x 40 and 5/8" long)
Diamond braided rope, preferably in light color to easily show ticks (3/8" x 100")
PVC pipe (3/4" diameter x 1m length)
All-purpose stainless-steel blade scissors (8")
Yard stick and/or measuring tape
Thread and sewing machine (recommended)

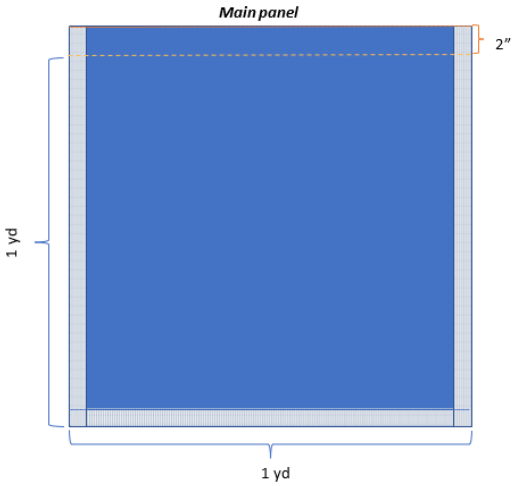
Construction:

1. Gather all supplies.
2. Measure and cut the fabric to 37 and 1/4" wide x 40 and 5/8" long (Panel-1).
3. To increase the durability of the tick drag, fold the panel at the crosshatched sections (Panel-1) and sew a (5/8") hem on each side except for the top. The resulting dimensions of the panel should be 1 yd square (Panel 2).

Panel-1:

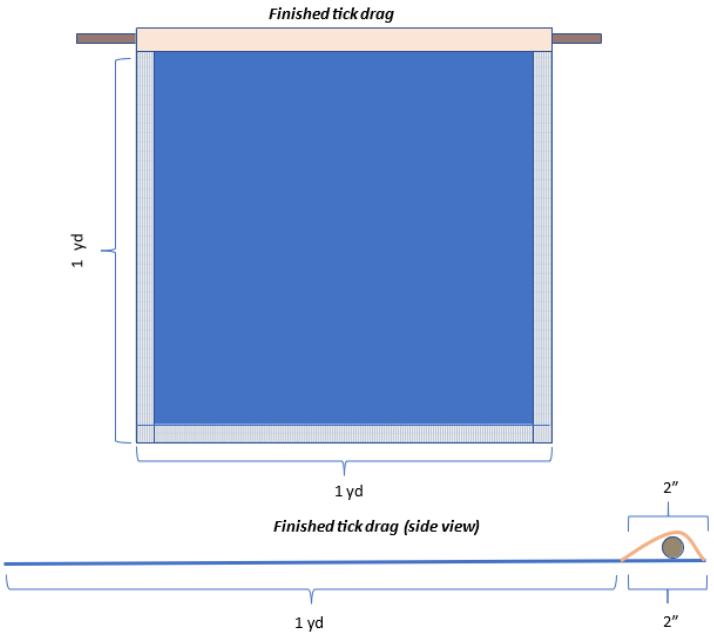


Panel-2:



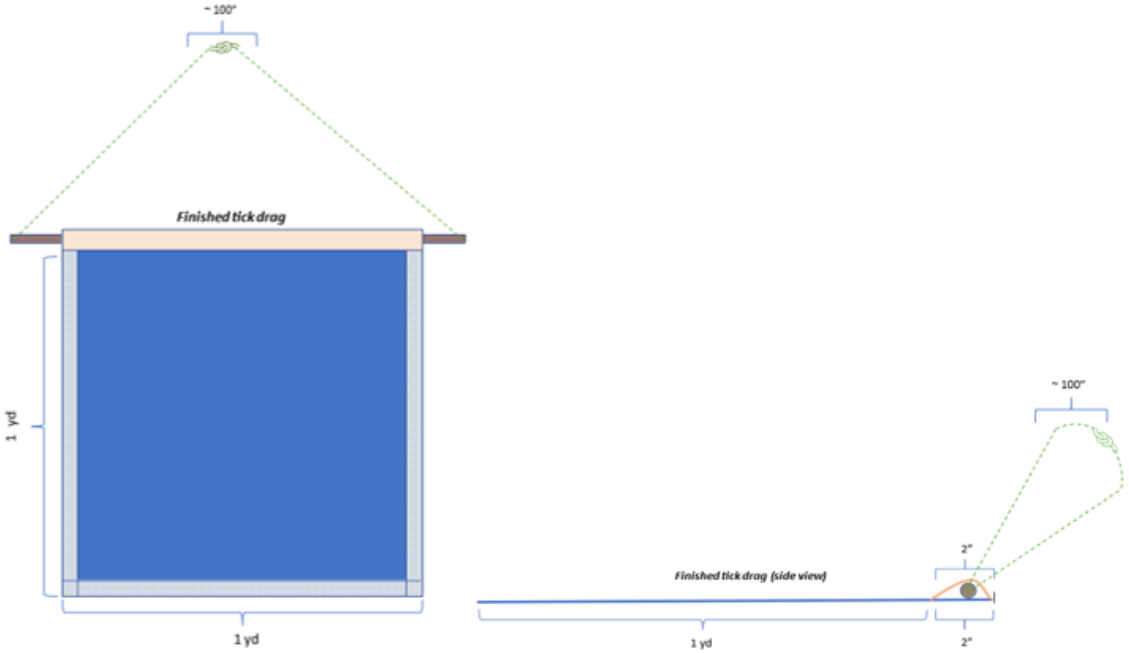
4. Fold the top of the fabric at 2" from the edge (Panel-2 at the orange dashed line). Sew along the bottom edge of the folded fabric leaving the sides open.
5. Insert the ¾" diameter PVC pipe through the top of the fabric (Panel-3).

Panel-3



6. Thread ~100" of braided rope through PVC pipe and tie ends securely Panel-4).

Panel-4


~ 100"
Finished tick drag
1 yd
1 yd
~ 100"
2"
Finished tick drag (side view)
1 yd
2"

Appendix D: Tick Flag Construction

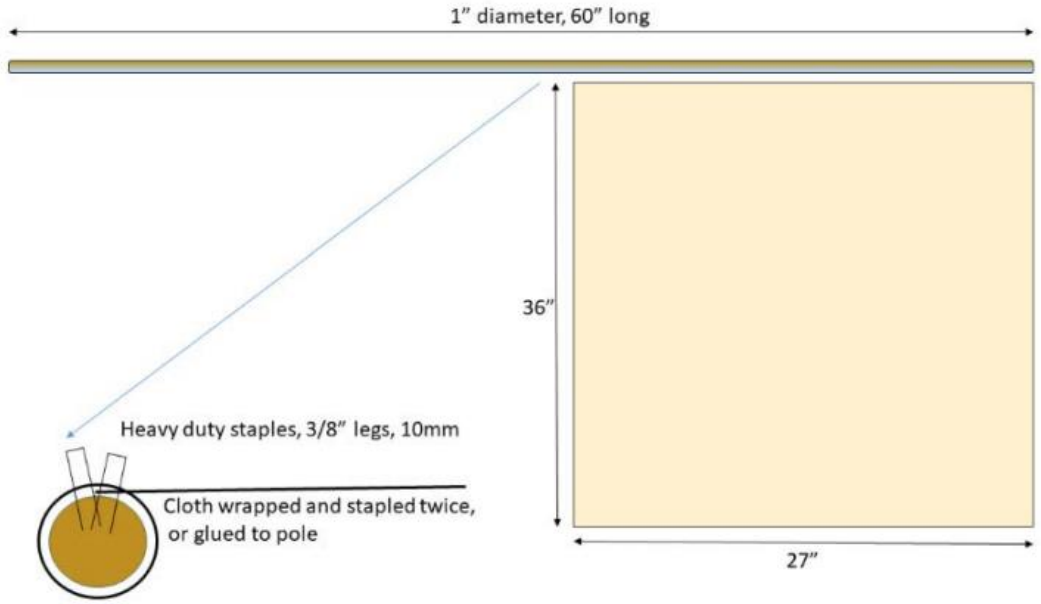
Adapted from: Centers for Disease Control and Prevention (CDC). (2020). Guide to the surveillance of metastriate ticks (Acari: Ixodidae) and their pathogens in the United States. *Division of Vector-Borne Diseases, CDC, Atlanta, GA and Ft. Collins, CO. https://www. cdc. gov/ticks/pdfs/Tick_ surveillance-P. pdf*, 49.

Materials
1” diameter by 60” long wooden dowel
Heavy duty stapler
Heavy duty 3/8” staples
Crib Cloth, flannel, or corduroy (approx. 27” x 36”)
Hammer

Construction:

1. Cut cloth to the appropriate size (sewing or hemming will not be required if the laminated fabric is used)
2. Staple the shorter edge of the cloth to broom stick at one end along the whole edge of the cloth
3. Hammer staples down if needed so they are flat on the broomstick
4. Wrap cloth around broomstick until the cloth overlaps, staple again beside the previous layer’s staples
5. Hammer staples down if needed so they are flat on the broomstick

Assembly diagram:



Appendix E: Tick Sweep Construction

Materials
(1) 10’ x ¾” PVC40 Pipe
(2) ¾” PVC40 Cap SLIP (Fig 1)
(3) ¾” PVC40 Male Adapter MPT x SLIP
(2) ¾” PVC40 Female Adapter SLIP x FPT (Fig 3)
(1) ¾” PVC40 45° Elbow SLIP x SLIP (Fig 4)
(1) ¾” PVC40 Riser Extender FPT x MPT (Fig 4)
(1) Double-sided waterproof flannel
(1) 8oz PVC Cement (Regular)

[1] Available at www.amazon.com Item is called: Flannel Crib Protector Pad, 100% Waterproof Mattress Protector Pad, One Size, 27” X 50”

Construction:

1. Cut 10’ PVC pipe into six 18” sections (One 10’ length makes 1.5 flags)
2. Section A - Cement (1) PVC Cap Slip and (1) ¾” Male Adapter on opposing ends of one 18” piece (Fig 5A)
3. Section B - Cement (1) ¾” Male Adapter and (1) ¾” female Adapter on opposing ends of one 18” piece (Fig 5B and 5C)
4. Section C - Repeat Step 3 making two identical pieces
5. Section D - Cement (1) ¾” PVC Cap Slip and (1) ¾” 45° Elbow on opposing ends of one 18” piece (Fig 5D)
6. Cement (1) ¾” PVC40 Riser Extender into the 45° Elbow of Section D
7. Measure and cut the flannel material into a rectangle of 16” x 24” (One yard makes approximately four sweeps)
8. Using heavy-duty masking tape or duct tape, attach the 16” side of the flannel material to Section D. Position the material so that the attachment point is at the bottom of the sweep when held upright. Reinforce with two layers of tape if using masking tape (Fig 6)
9. Let the cement dry per labeled instructions and assemble

NOTE: This design allows for the flag to be collapsible for easy packing and carrying.

Fig 1. ¾” PVC40 Cap SLIP

Fig 2. ¾” PVC40 Male Adapter MPT x SLIP

Fig 3. ¾” PVC40 Female Adapter SLIP x FPT

Fig 4. ¾” PVC40 Riser Extender FPT x MPT

Fig 5. The four completed sections labeled.

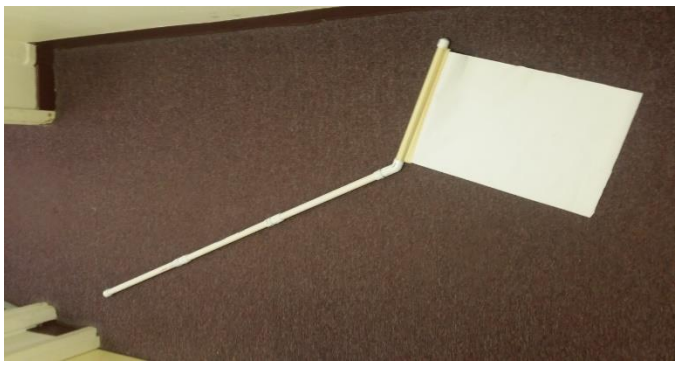


Fig 6. The four completed sections labeled.

Appendix F: CO_2 Trap Construction

Supplies (estimated prices as of 2021):

Materials
(1) ~14 Quarts Styrofoam Cooler
(1) Fiberboard Base or Plywood Base
145 cm Double-Sided 3M Carpet Tape
1 ± 0.09 kg Dry Ice (lasts for 24 hrs., may use less if deploying for shorter duration)

Note: Either fiberboard or plywood works, although plywood may be more durable.

Construction:

1. Drill two ~0.2” holes at the bottom of each side of the cooler.
2. Place double-sided carpet tape along the top edges of the fiberboard base.
3. Place enough dry ice to last the duration of the collection period.

Appendix G: Field Data Collection Form

Field Data Collection Form

COLLECTION INFORMATION

Collecting Agency: Contact Name: ______________________________ Name of Agency: ______________________________	**Tick Collection Date (MM/DD/YYYY)** ____/____/_______

SITE INFORMATION

Surveillance Site Name	**Transect Number**	**Street Address**	**City/State**	**County**
Surveillance Site Latitude (##.######)		**Surveillance Site Longitude (##.######)**		
Habitat Type ☐ Brush ☐ Forest ☐ Grassland ☐ Mixed forest and brush ☐ Mixed forest and grassland ☐ Mixed grassland and brush ☐ Not determined ☐ Other: ______________________		**Site Ownership Type** ☐ Private property ☐ Public land		
Start time ____:____ ☐ AM ☐ PM	**End time:** ____:____ ☐ AM ☐ PM	**Density sampling method** ☐Distance dragging ☐Distance flagging	**Exploratory sampling (if used)** ☐Tick dragging ☐ Tick flagging ☐ Tick sweep ☐ CO_2 trap	
# of m² Dragged/Flagged	**Additional comments:**		**Problems?** ☐Yes ☐ No	
CO_2 Trap Deployment Time				

WEATHER INFORMATION (<u>Complete on the field</u>)

Temperature	**Relative humidity**	**Wind speed**
_______ ☐ °F ☐ ℃ ☐ K	_______ %	_______ ☐ mph ☐ km/h ☐ mps ☐ m/s ☐ ft/s

COMPLETE COLLECTION TABLE BELOW WITH FINDINGS: (<u>May be completed after field collection</u>)

Tick identified	# adult male	# adult female	# adult unknown	# nymphs	Larvae collected?
Genus: **Species:**	**Total:** **Fed:** **Unfed:**	**Total:** **Fed:** **Unfed:**	**Total:** **Fed:** **Unfed:**	**Total:** **Fed:** **Unfed:**	☐ Yes ☐ No **If yes, how many?** ________
Genus: **Species:**	**Total:** **Fed:** **Unfed:**	**Total:** **Fed:** **Unfed:**	**Total:** **Fed:** **Unfed:**	**Total:** **Fed:** **Unfed:**	☐ Yes ☐ No **If yes, how many?** ________
Genus: **Species:**	**Total:** **Fed:** **Unfed:**	**Total:** **Fed:** **Unfed:**	**Total:** **Fed:** **Unfed:**	**Total:** **Fed:** **Unfed:**	☐ Yes ☐ No **If yes, how many?** ________
Genus: **Species:**	**Total:** **Fed:** **Unfed:**	**Total:** **Fed:** **Unfed:**	**Total:** **Fed:** **Unfed:**	**Total:** **Fed:** **Unfed:**	☐ Yes ☐ No **If yes, how many?** ________

Appendix H: Tick Identification Guides

Durden, L. A., & Keirans, J. E. (1996). Nymphs of the genus Ixodes (Acari: Ixodidae) of the United States: taxonomy, identification key, distribution, hosts, and medical/veterinary importance. Entomological Society of America.*

Keirans, J. E., & Clifford, C. M. (1978). The genus Ixodes in the United States: a scanning electron microscope study and key to the adults. Journal of medical entomology. Supplement, 2, 1–149. https://doi.org/10.1093/jmedent/15.suppl2.1

Yunker, C. E., Keirans, J. E., Clifford, C. M., & Easton, E. R. (1986). Dermacentor ticks (Acari: Ixodoidae: Ixodidae) of the new world: a scanning electron microscope atlas. Proceedings of the Entomological Society of Washington, 88(4), 609-627.

Keirans, J. E., & Litwak, T. R. (1989). Pictorial key to the adults of hard ticks, family Ixodidae (Ixodida: Ixodoidea), east of the Mississippi River. Journal of medical entomology, 26(5), 435–448. https://doi.org/10.1093/jmedent/26.5.435

Keirans, J. E., & Durden, L. A. (1998). Illustrated key to nymphs of the tick genus Amblyomma (Acari: Ixodidae) found in the United States. Journal of medical entomology, 35(4), 489–495. https://doi.org/10.1093/jmedent/35.4.489

Egizi, A. M., Robbins, R. G., Beati, L., Nava, S., Vans, C. R., Occi, J. L., & Fonseca, D. M. (2019). A pictorial key to differentiate the recently detected exotic Haemaphysalis longicornis Neumann, 1901 (Acari, Ixodidae) from native congeners in North America. ZooKeys, (818), 117–128. https://doi.org/10.3897/zookeys.818.30448

Tutorial for tick identification (click "Interactive Program for Teaching Tick Morphology"):

https://www.acq.osd.mil/eie/afpmb/training_resources.html

*Durden & Keirans (1996) available upon request. Please email cdsvectorteam@doh.nj.gov to obtain a copy.

Appendix I: Voucher Instructions

If a novel species is detected, agencies should place specimens from each collection in microfuge tubes with ~80% ethanol (undenatured or denatured). Do not mix specimens from different days or locations in the same vial.

Shipping specimens for PHEL identification:
James Occi
Microbiologist
NJDOH Public Health and Environmental Laboratories (PHEL)
Lab L-315
3 Schwartzkopf Drive
Ewing, NJ 08628
E-mail: james.occi@doh.nj.gov

Please include the form below. Include one sheet per collection. Add as many fields as necessary.

Novel Specimen Voucher Form

Collector	Date of collection	Collection State	Collection County	Surveillance Site Latitude (##.######)	Surveillance Site Longitude (##.######)	Removed from animal?
						☐ Yes ☐ No If yes, animal species: _____
						☐ Yes ☐ No If yes, animal species: _____

Also print out a label (laser not inkjet) and insert in each vial. Label samples like below:

Ixodes scapularis
Watchung Res.
County Name., NJ
Date/Collector

Ixodes scapularis
Watchung Res.
Union Co., NJ
7April2025/ J. Occi

Appendix J: Calculating Tick Density and Infection Prevalence

County Status Classification:

Established: ≥ 6 ticks of a single life stage or > 1 life stage collected per county withing a 12-month period.

Reported: < 6 ticks of a single life stage collected per county within a 12-month period.

No records (note: should not be interpreted as absence or occurrence).

Density of nymphs (DON): total no. of nymphs collected / total area sampled (usually expressed in 100 m^2)

Density of females (DOF): total no. of adult females collected / total area sampled (usually expressed in 100 m^2)

Nymph Infection Prevalence: no. of infected nymphs / total number of nymphs tested

Adult Female Infection Prevalence: no. of infected adult females / total number of adult females tested

Density of host-seeking infected nymphs (DIN or Entomological Risk Index): DON x Nymph Infection Prevalence

Density of host-seeking infected adult female ticks (DIF): DOF x Adult Female Infection Prevalence

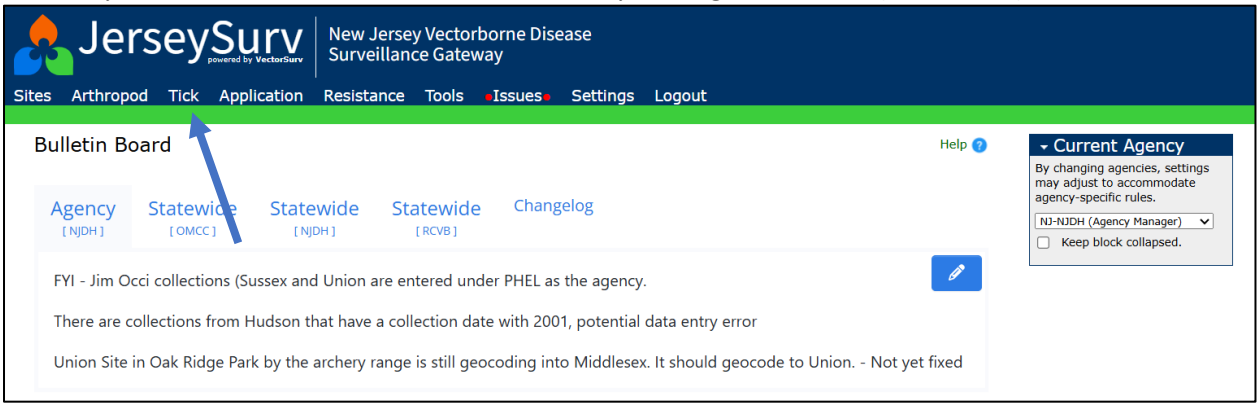
Minimum Infection Rate: [no. of positive pools / total specimens tested] x 1000

Appendix K: JerseySurv Tick Module Entry

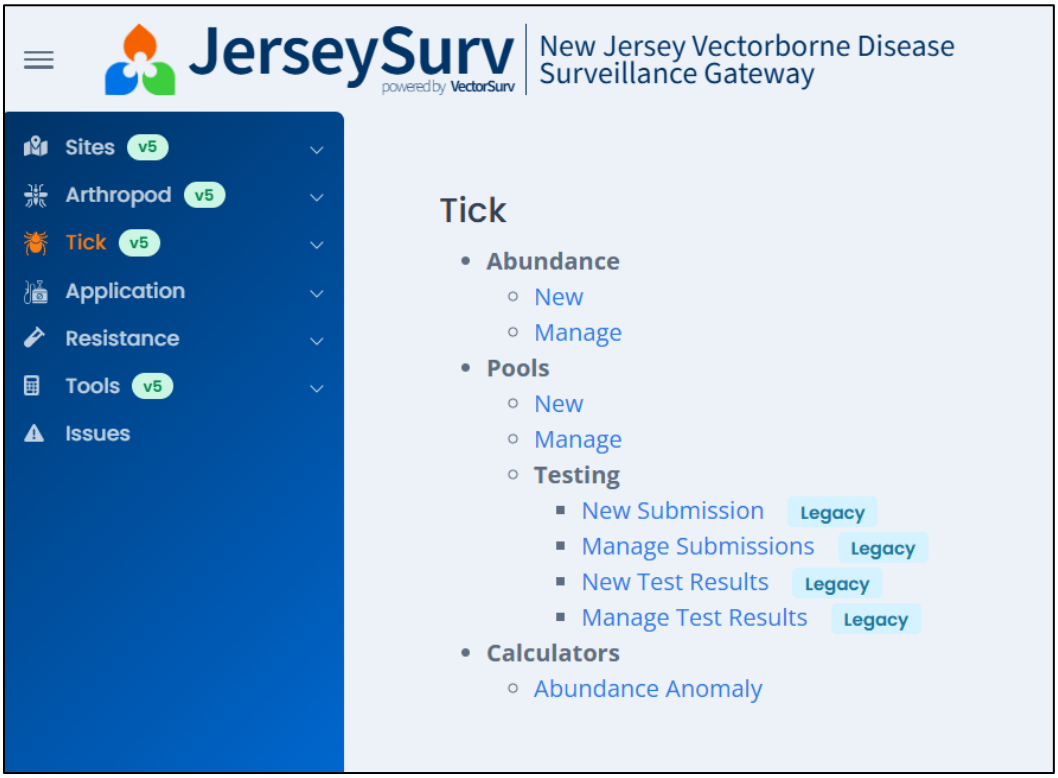
NOTE: Tick collection data must be entered first before pool data. Create a **Collection** first and then link **Pools** to their respective collection. A video tutorial for creating collections and pools is located here.

Accessing The Tick Module

1. On JerseySurv, tick collection data can be entered by clicking the **Tick** tab (blue arrow).



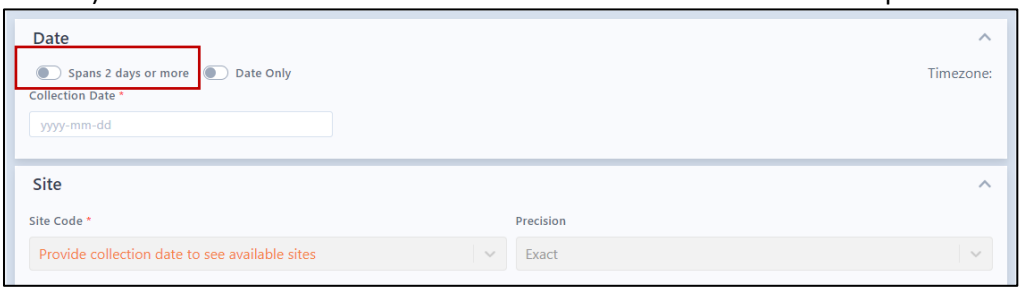
2. New abundance and pool data can be entered through their respective sections.



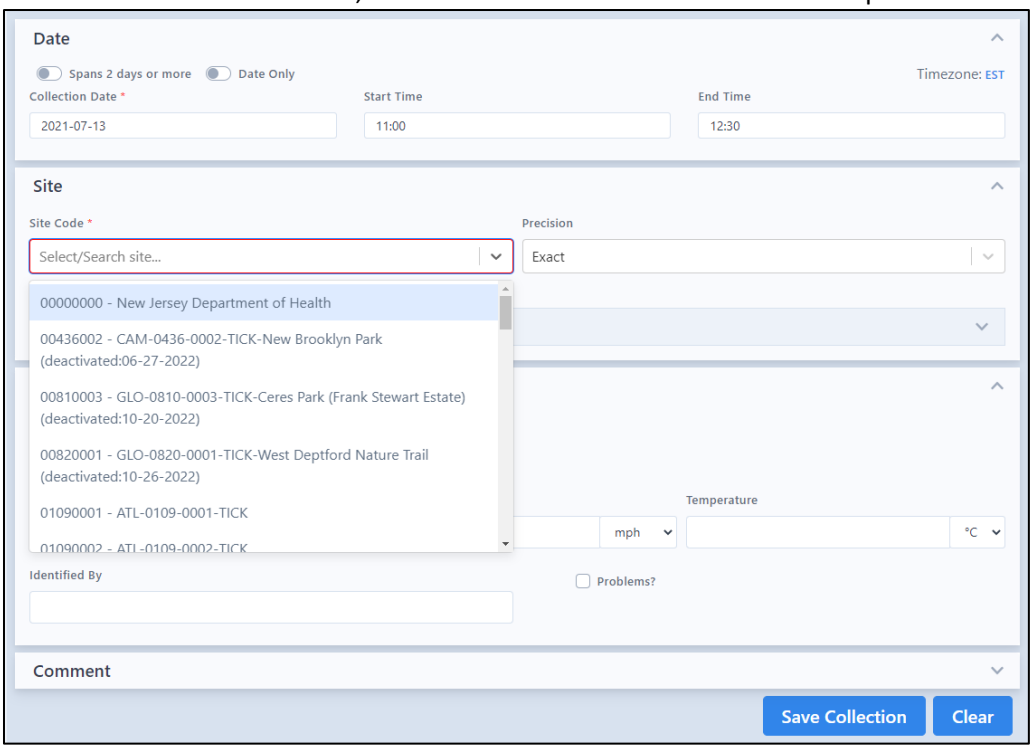
Creating A New Collection

3. After clicking **New** under the **Abundance** section, information related to new collections can be

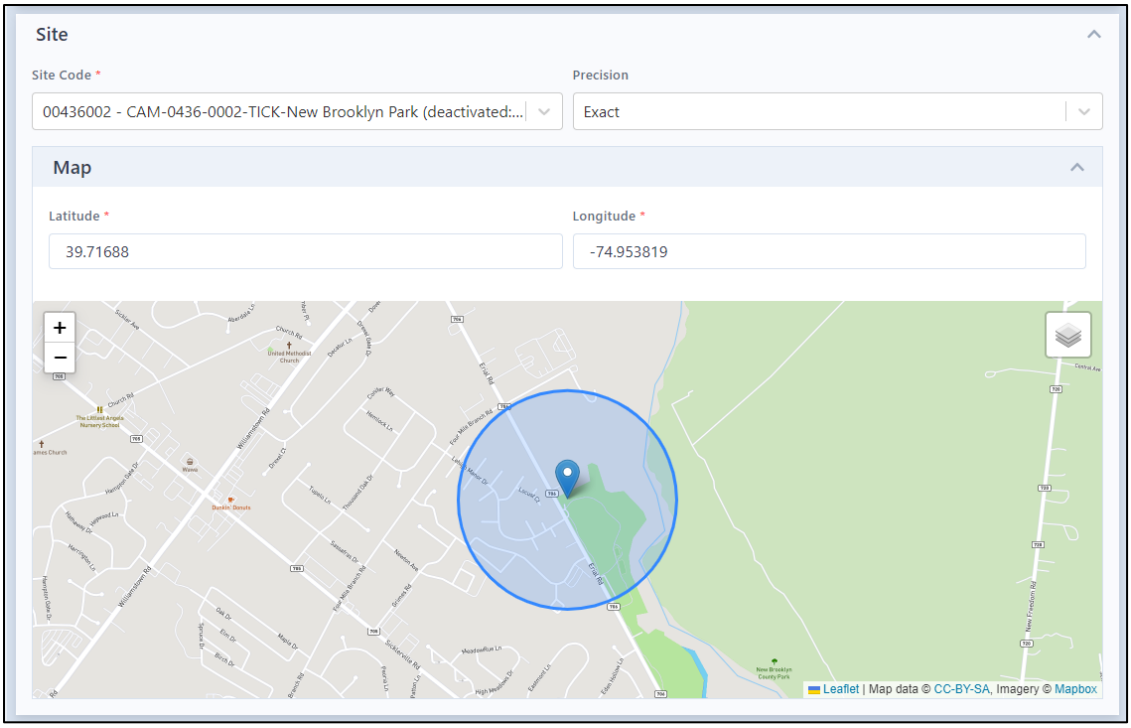
entered. First, enter the collection date, and then the start/end times. **Ensure that the timezone is correct.** If collection spans two days or more, toggle the switch labelled **Spans 2 days or more** (in the red box). Note: **Site Code** would not be available if **Collection Date** is not specified.



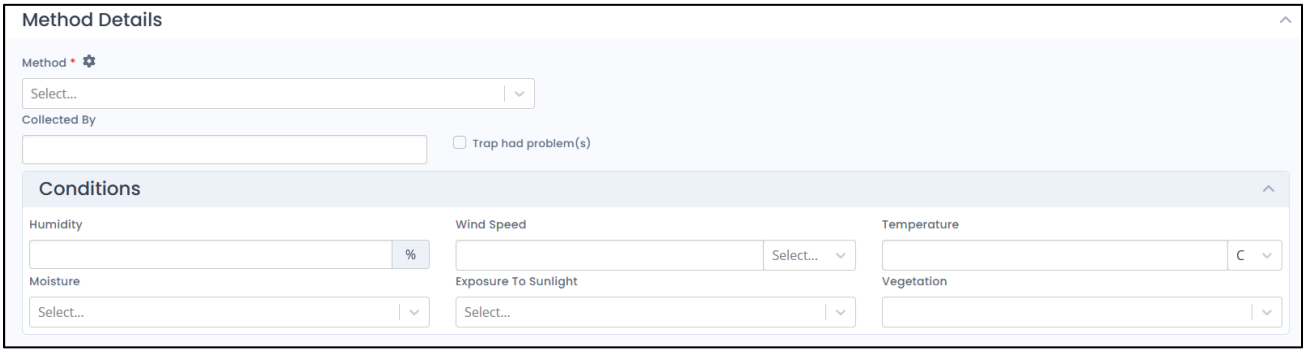
4. After the **Date** section is filled, **Site Codes** will be available under the drop-down menu.



5. Clicking the **Map** tab allows you to view and review where the collection was conducted.



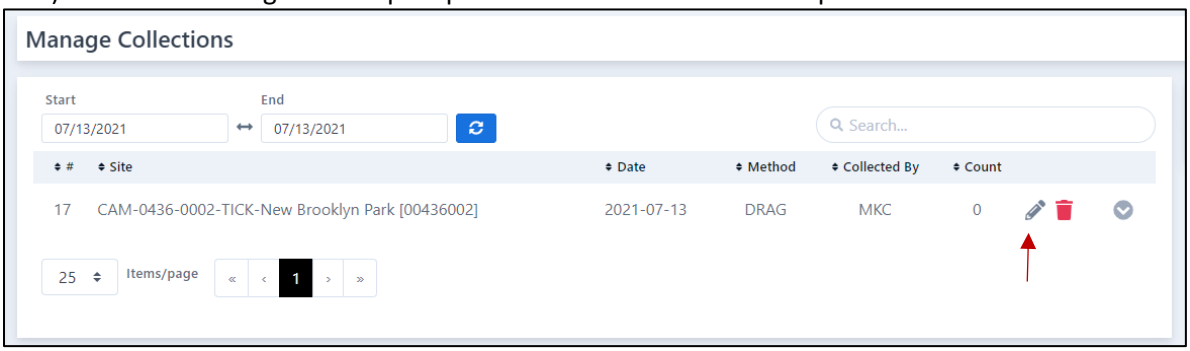
6. Under the **Method Details** section, the tick collection method as well as environmental parameters from the collection can be inputted. For tick dragging (DRAG) and flagging (FLAG) methods, the density sampling method and the amount of area sampled can be inputted in their respective fields. If any problems arise during the collection, the investigator can check the **Trap had problem(s)?** checkbox and place a comment under the **Comment** section.



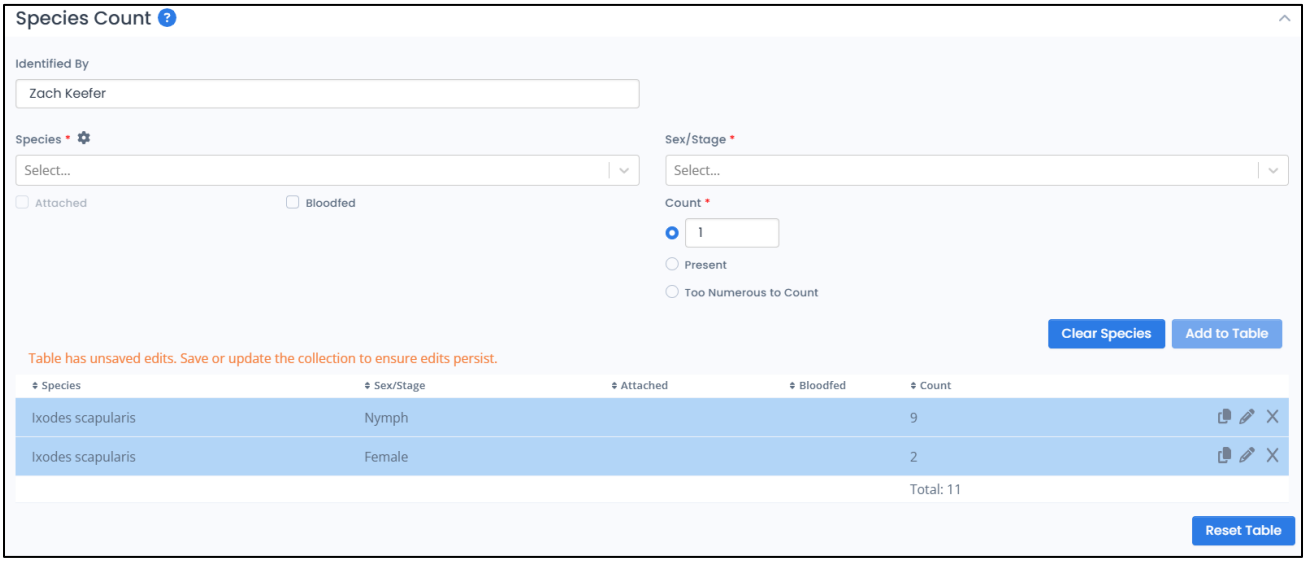
7. After the **Date**, **Site**, **Method Details**, and **Comment** sections are completed, press **Save Collection** to create a new collection.

Managing Tick Collections

8. Collections created can be accessed by clicking **Abundance > Manage**. Clicking the edit button (pencil icon) allows an investigator to input species count and create or link pool data.

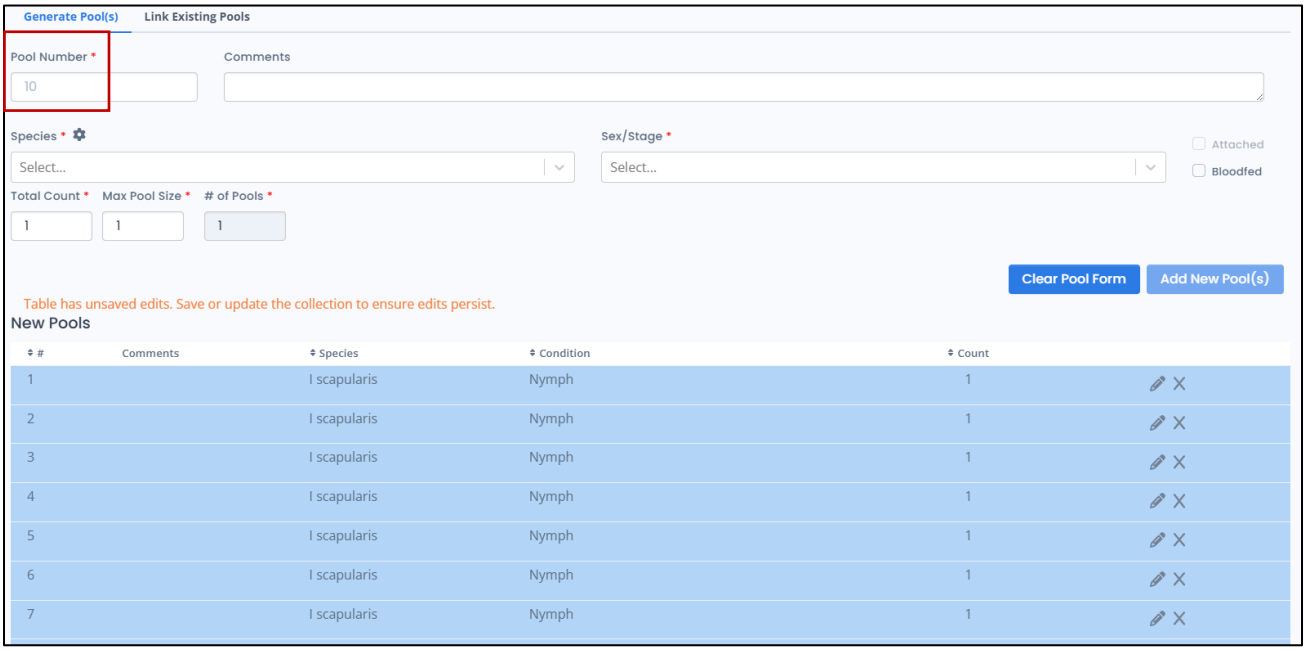


9. Under **Species Count**, abundance data can be inputted based on species, sex (only adults should be sexed), and life stage. New species or life stages can be added by clicking the **Add to table** button.

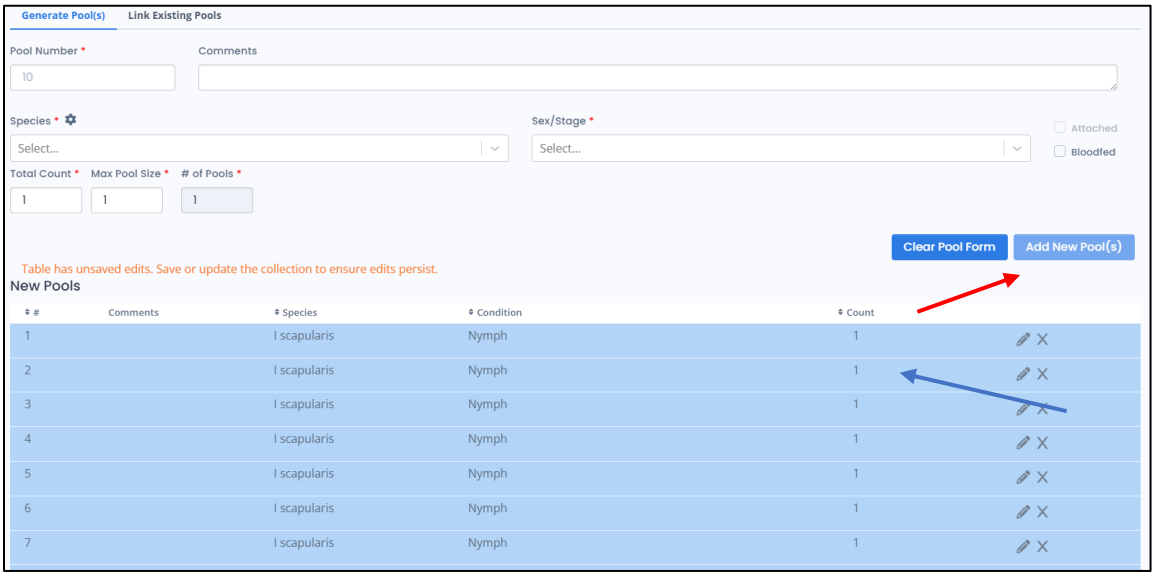


Creating New Pools from the Collection Page

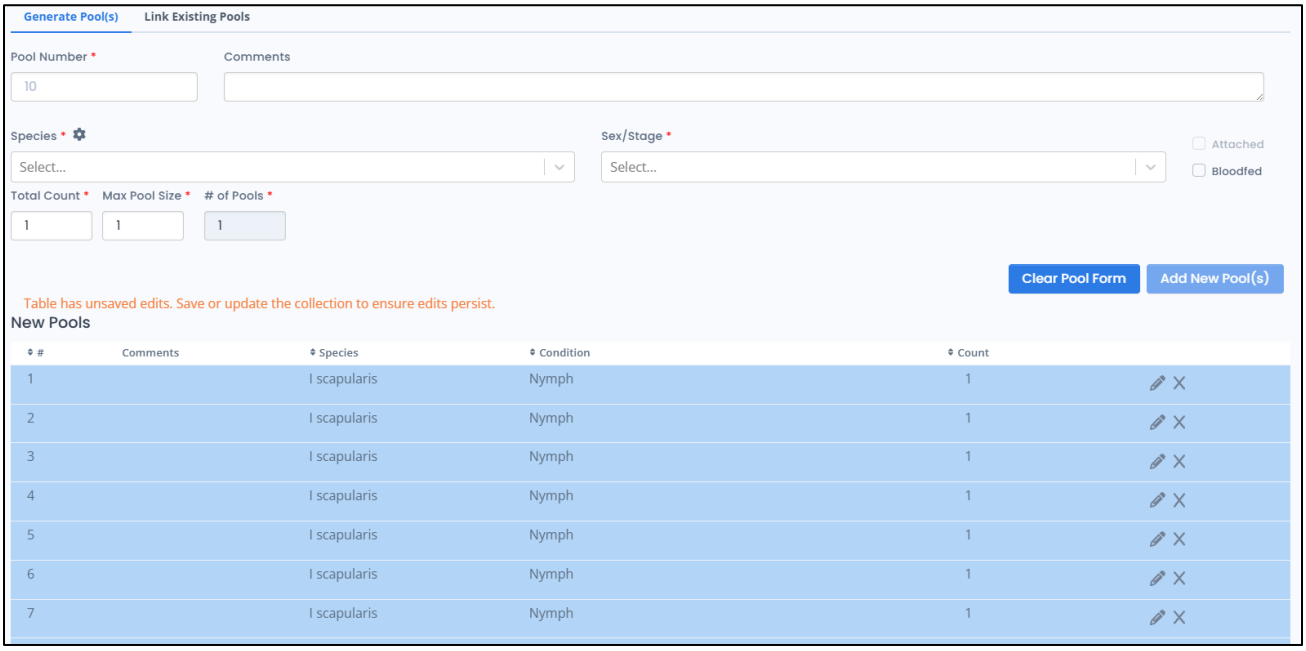
10. New tick pools can be created and linked simultaneously at the bottom of the Collection page after navigating to **Abundance > Manage > Collection**. The investigator should generate pool numbers starting from 1, or whatever is recommended (red box). **However, because pool numbers account for both mosquito and tick pools, it is recommended to set the tick pool numbers to something above the usual number of mosquito pools submitted (ex., 5000), so that tick pools can be grouped together.** Species, stage, sex, count, and other information can be inputted in their respective fields.



11. Once the required fields are filled, the pool can be generated by clicking the **Add New Pool(s)** button (red arrow). Pools generated will show up at the bottom of the page (blue arrow). Pools can be edited and unlinked to the collection if desired.



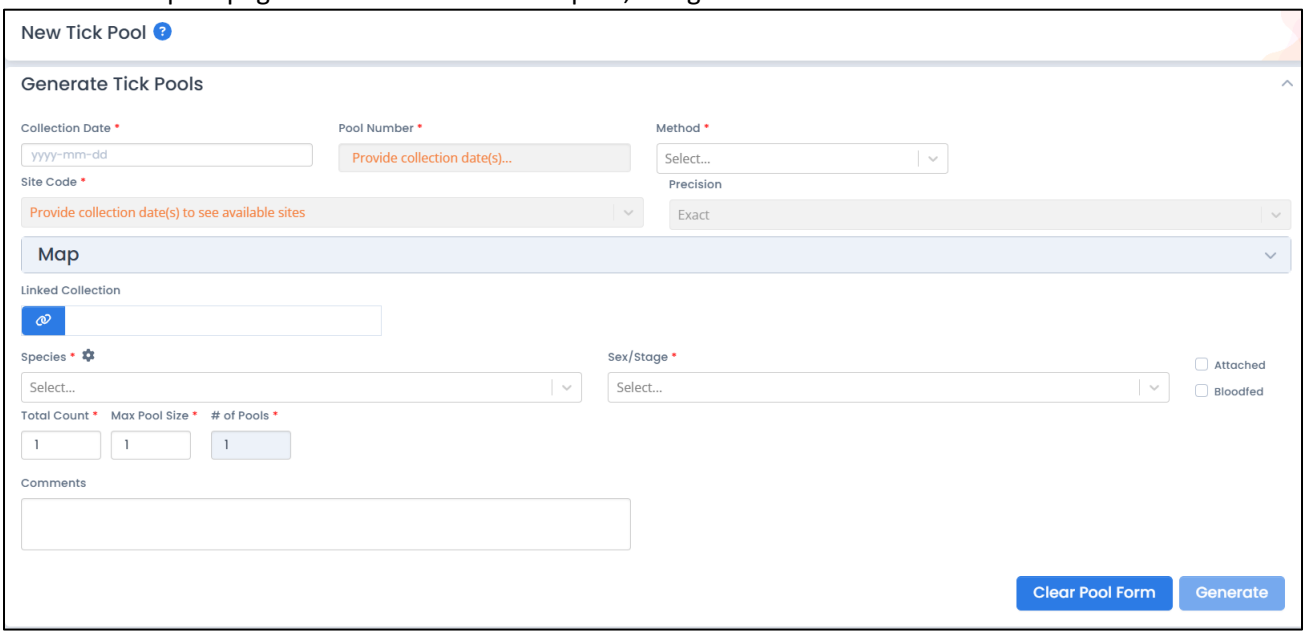
12. In cases where individual ticks are tested rather than pooled, the number of **# of Pools** should be equal to the number of specimens tested and **Max Pool Size** should be 1. Under the **Total Count** box, a message appears to indicate to the investigator that pool numbers will be generated.



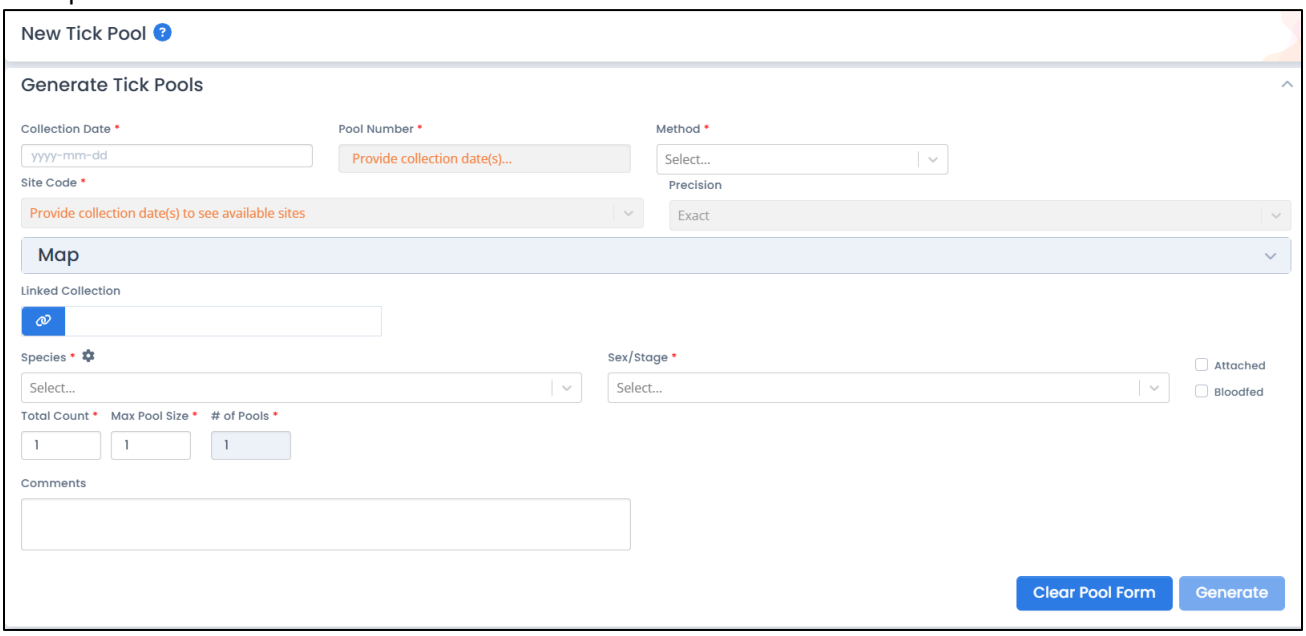
13. After clicking **Add New Pool(s)**, four new pools will be generated. Pools can be edited and unlinked to the collection as desired.

Creating New Pools from the Pools Page

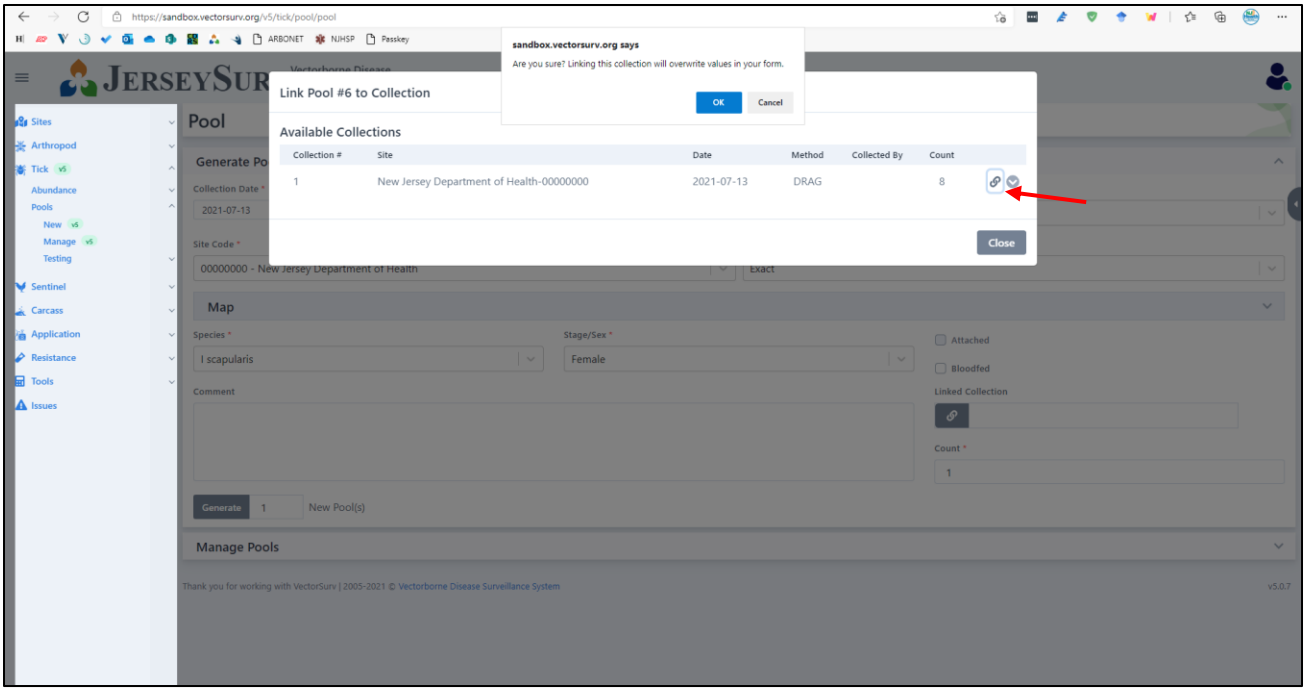
14. To access the pool page and create an unlinked pool, navigate to **Pools > New**.



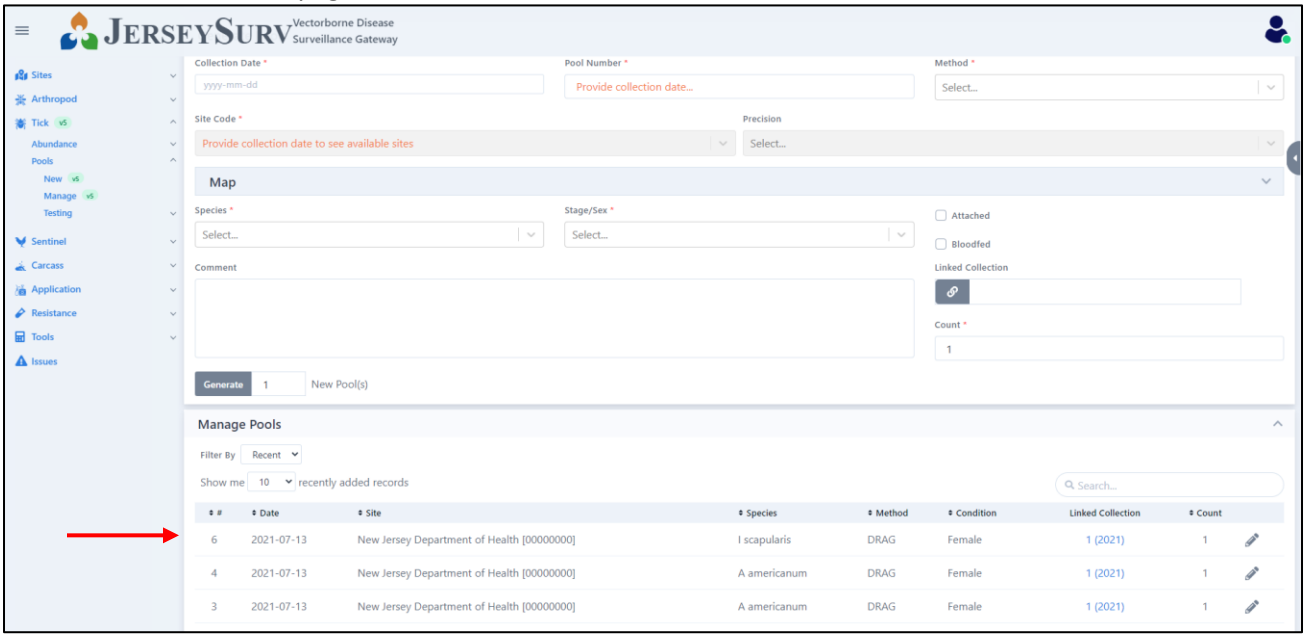
15. **Collection Date** needs to be filled out before **Pool Number** can be created. Fill out all the required and option fields as needed.



16. Clicking the **Linked Collection** button will display the collections that the pool can be linked to. Clicking the link button (red arrow) and selecting **OK** will allow the investigator to link the pool to the selected collection. Note: as displayed on the message, linking a collection will overwrite some values in the form.

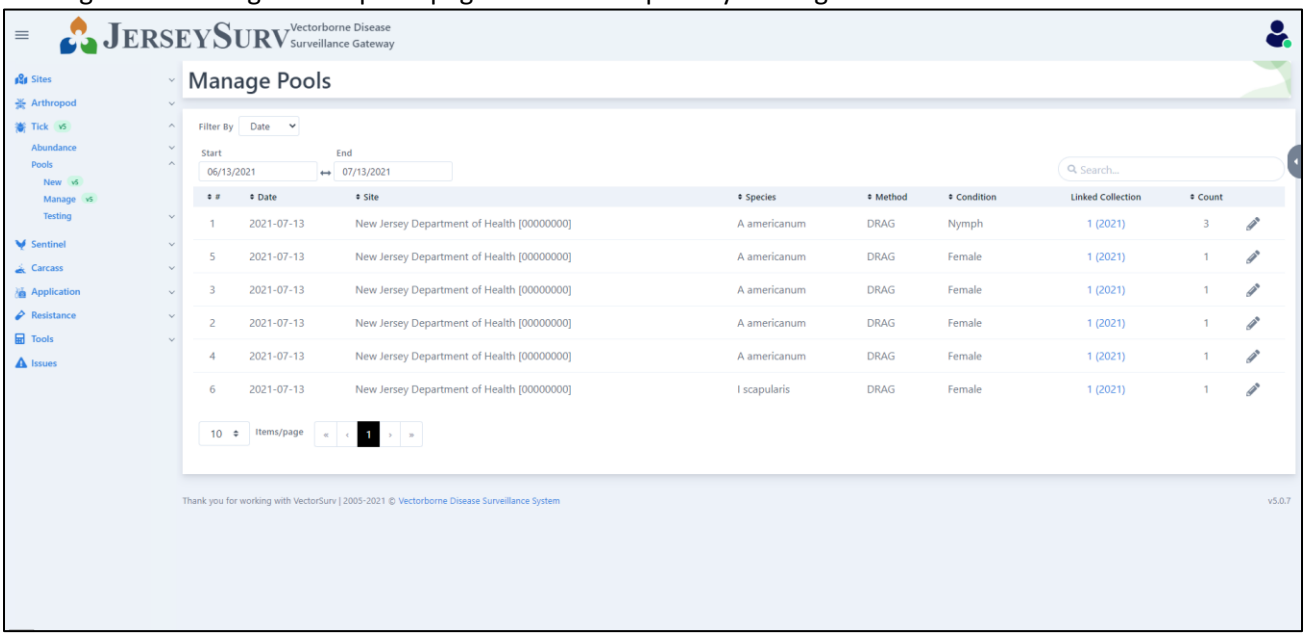


17. After linking and clicking **Generate**, the new pool (red arrow) will be available under the **Manage Pools** tab at the bottom of the page.



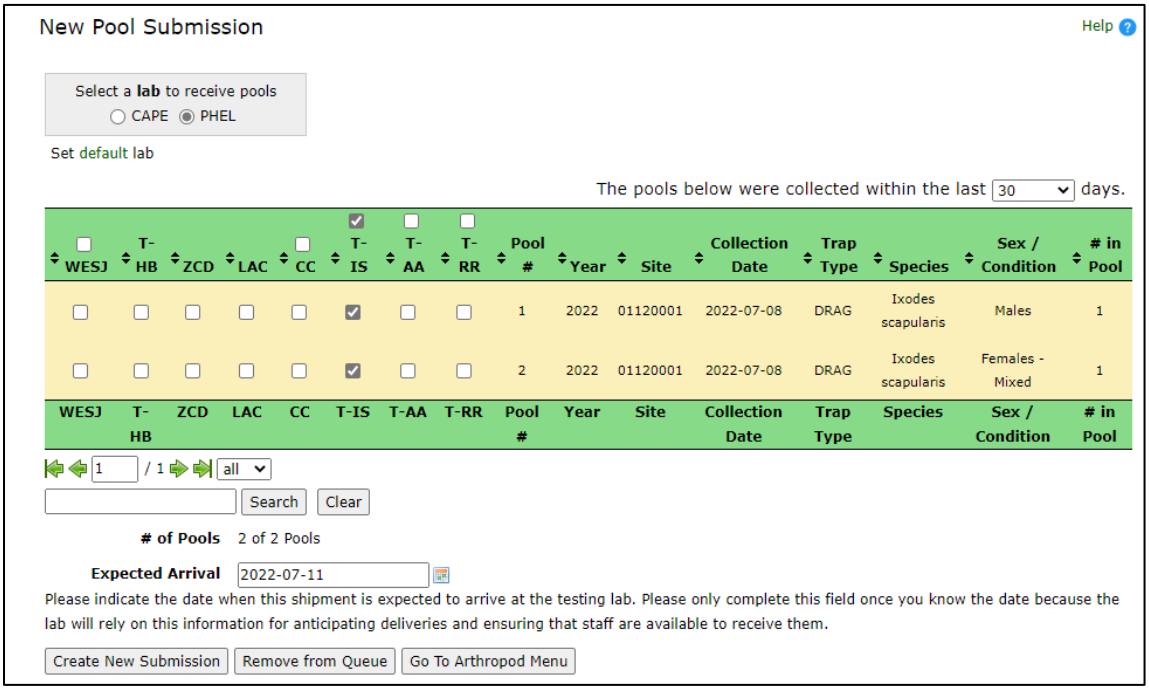
Managing Pools

18. Existing pools can be viewed and managed by navigating to **Pools > Manage**. On this page, the investigator can navigate to a pools page and edit the pool by clicking the icon.

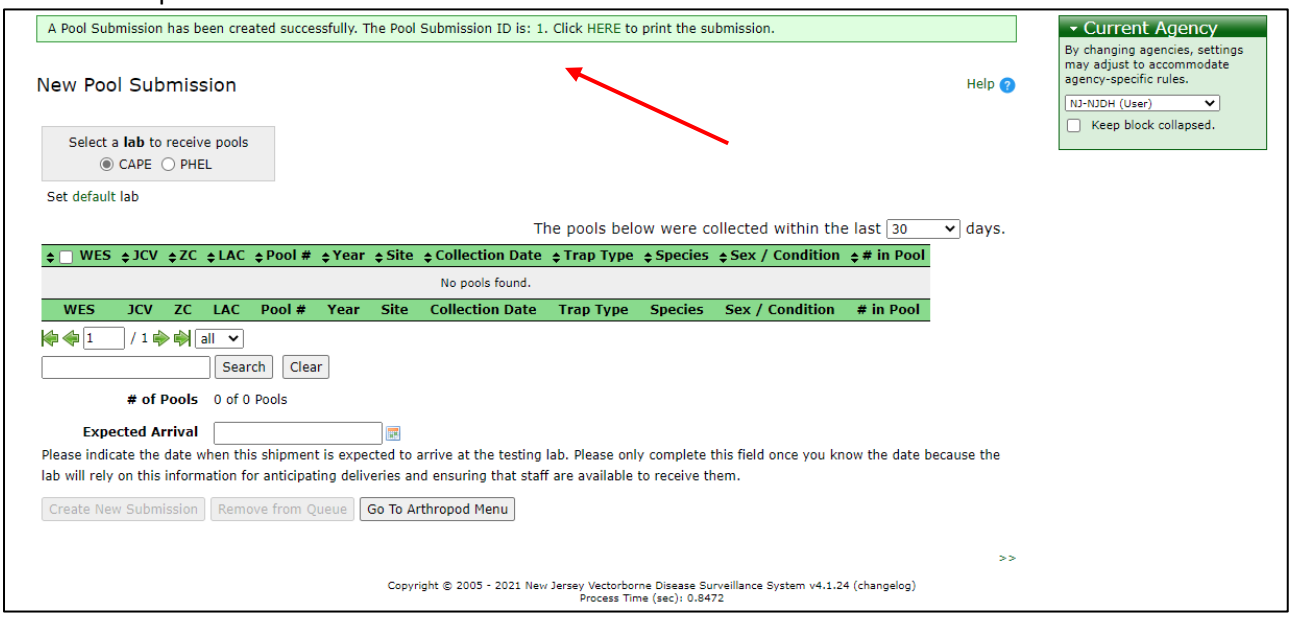


Creating New Pool Submissions for Testing

19. To create a new pool submission for testing, from the **Tick Module**, navigate to **Pools > Testing > New Submission**. From the **New Pool Submission** page, first click the lab that the samples should be sent to (currently, ticks are to be sent to NJPHEL). Then, indicate the type of tests to be performed for each pool. Then, input the date of the **Expected Arrival**. Finally, click the **Create New Submission** button.

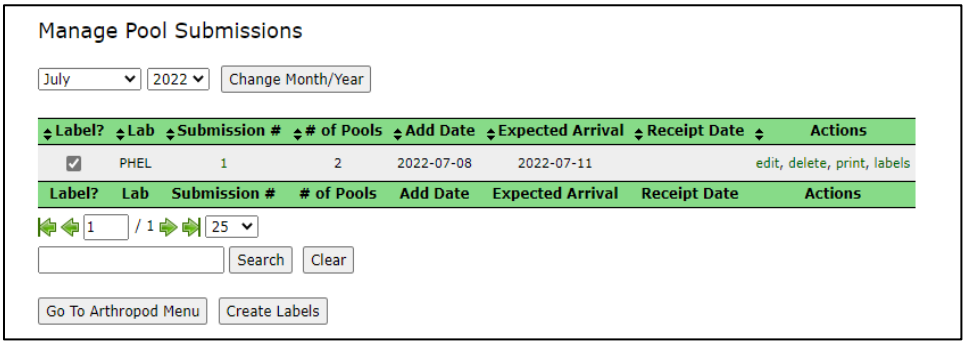


20. After the pool has been submitted, a message at the top (red arrow) will display indicating successful creation of a pool submission as well as the Pool Submission ID.

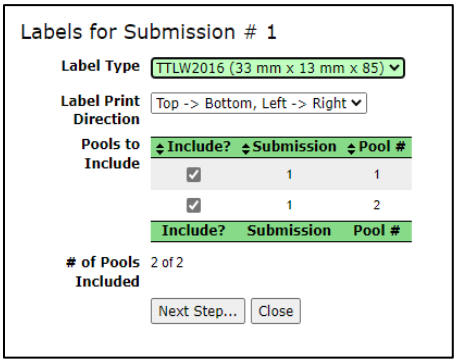


Managing Pool Submissions and Printing Labels

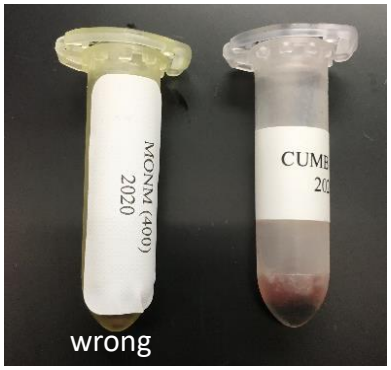
21. Newly submitted pools can be managed by clicking the Tick tab and navigating to **Pools > Testing > Manage Submissions**. In the **Manage Pool Submissions** page, the investigator may edit, delete, print, or create labels for the pool submissions.



22. Clicking **print** will open a pop-up window that will include information related to the pools submitted.

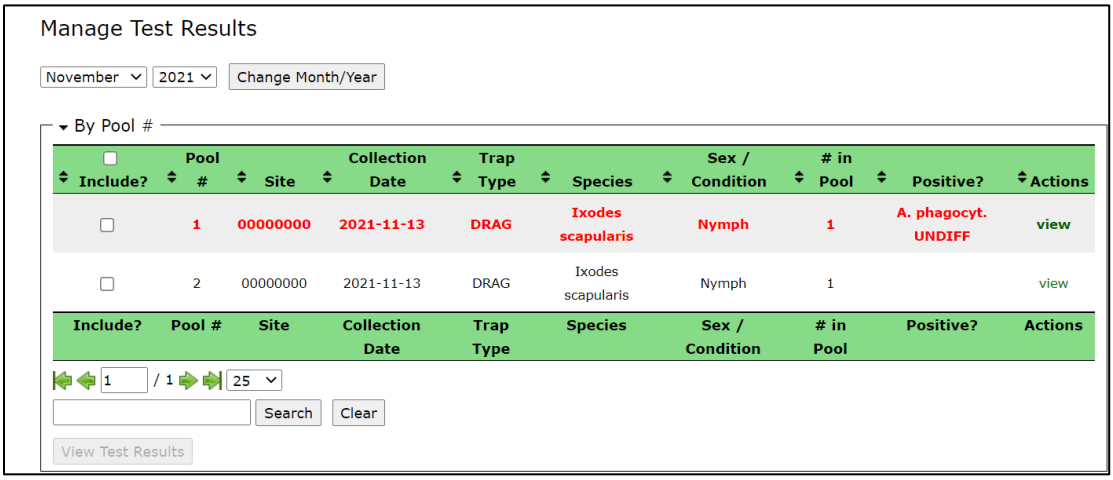


23. Clicking **labels** will open a window for label creation. After selecting the **Label Type**, **Label Print Direction**, and **Pools to Include**, clicking **Next Step…** will show a preview of how the printed label sheet looks like.
    a. Depending on the sheet this will be printed on, the label type may vary. For 16 x 4 labels on 8.5 x 11 paper (printer paper), **label type** will be "TTLW2016."
    b. Labels should be used for the side of the tube (see below).



24. After clicking **Next Step…** once again, a .zip folder will be generated containing the labels. Print and put the labels in the appropriate vials.

25. Results can be accessed by navigating to **Arthropod > Pools > Testing > Manage Test Results.**



26. After clicking **Manage Test Results**, you can view testing results.