Animal and Plant Health Inspection Service
U.S. DEPARTMENT OF AGRICULTURE

Monitoring *Haemaphysalis longicornis*, the Asian longhorned tick, populations in the United States

January 2024

Contents

Purpose

This document is a resource for United States Department of Agriculture, Animal and Plant Inspection Services, Veterinary Services (USDA-APHIS-VS) employees and our State and local partners to guide activities related to discovery of exotic *Haemaphysalis longicornis*, the Asian longhorned tick (ALHT)/bush tick/scrub tick, in a State or region of the United States (U.S.).

Goals for monitoring within a state or region include:

1) Initial detection of the tick;
2) Characterizing its distribution, habitat, and host associations;
3) Detecting severe tick infestations causing morbidity or mortality in animal populations; and
4) Correlating the presence of this tick with a disease agent that may be harmful to livestock or equine populations.

Information regarding diagnosis and reporting of foreign animal diseases (FAD) potentially associated with this tick is described elsewhere and is not included here. This guide also serves as a document of transparency for other national agencies that may want to develop similar or parallel plans for monitoring ALHTs in the United States. Overall, this specific guidance on surveillance and data management will improve our ability to monitor the spread of this tick and its impact on animal health.

Ecology and Etiology

Background

In November 2017, public health officials in Hunterdon County, New Jersey, reported the first detection of *H. longicornis* in the United States, outside of a port of entry inspection site. The presence of ALHT, a species not native to the United States, has since been confirmed in at least eighteen additional states, and based on current research studies, there were at least three separate introductions of this tick species into the United States, potentially from East Asia (Egizi et al. 2020). Retrospective studies also found an archival ALHT specimen collected at large in West Virginia as far back as 2010. As part of its mission to protect American agriculture, VS continues to provide updated information to stakeholders to nurture planning, surveillance, outreach, control, and overall collaboration for ALHT in the United States.

General Biology

The ALHT has a wide global distribution in temperate regions, such as Japan, Korea, China, Australia, and New Zealand. This tick species exhibits multiple reproductive types, including parthenogenetic and bisexual. Parthenogenesis allows for one female tick to create a population without mating and may quickly lead to massive infestations causing morbidity and mortality in livestock in its established range. The parthenogenetic type of the ALHT easily invades and establishes in new areas (e.g., Australia, New Zealand). Currently, all known U.S. ALHT populations are parthenogenetic.

In the United States, ALHT are currently reported in Arkansas, Connecticut, Delaware, Georgia, Indiana, Kentucky, Maryland, Massachusetts, Missouri, New Jersey, New York, North Carolina, Ohio, Pennsylvania, Rhode Island, South Carolina, Tennessee, Virginia, and West Virginia. Several habitat suitability models suggest that populations of this tick species could potentially inhabit most of the eastern half of the United States and areas on the Pacific Coastline (Magori 2018, Rochlin 2019, Namgyal et al. 2020). In areas outside of the United States, where it is established, the ALHT adapts to a wide range of environmental conditions, but invasive populations elsewhere prefer mild temperatures and humid climates. To survive colder temperatures, these ticks may enter diapause (suspended development) at any life stage (Heath 2016, Lawrence et al. 2017). In the United States, host-seeking nymphs are most active in the spring, adults in the summer, and larval stages in the fall. They may occur in a variety of habitat types, including fields, forest edges, meadows, and pastures (Thompson et al. 2021).

The ALHT is a three-host tick species. Three-host ticks normally seek a new individual animal host on which to feed for each of three active life stages, and then, after each blood meal, they return to the ground to either molt to the next stage or lay eggs. The ALHT has a wide host range. Unlike many other three-host tick species, earlier stages (larvae and nymphs) do not seem to prefer small mammals as hosts, and all three active life stages (larvae, nymphs, and adults) usually feed on the same medium- to large-sized host species. To date, the United States host list for the ALHT includes dogs, cats, cattle, goats, sheep, chickens, pigs, horses, white-tailed deer, elk, black bears, opossums, grey foxes, red foxes, coyotes, eastern cottontails, deer mice, groundhogs, raccoons, striped skunks, gray squirrels, eastern chipmunks, red-tailed hawks, great horned owls, barred owls, Canada geese, brown boobys, gray catbirds, blue jays, Carolina wrens, northern cardinals, house wrens, song sparrows, and humans (APHIS VS Situation Report, October 2023; Pandey et al. 2022).

The ALHT is a livestock pest that can transmit a variety of microbial pathogens, such as species of *Anaplasma, Babesia, Borrelia*, *Ehrlichia*, *Rickettsia*, and *Theileria* (Heath 2002, 2016; Multiple laboratory transmission studies with U.S. ALHT populations have shown that they are not competent vectors for *Borrelia burgdorferi* (Lyme disease), *Anaplasma phagocytophilum* (human granulocytic anaplasmosis), or *Franciscella tularensis* (tularemia); however, this tick species can transmit *Rickettsia rickettsii* (Rocky Mountain spotted fever), *Theileria orientalis* Ikeda (bovine theileriosis), Heartland virus, and Bourbon virus (Breuner et al. 2019, Levin et al. 2021, Tully and Huntley 2020, Stanley et al. 2020, Dinkel et al. 2021, Raney et al., 2022a; Raney et al. 2022b). *Theileria orientalis* Ikeda, *Theileria cervi, Babesia microti*, *Babesia* sp. Coco, *Babesia* sensu lato, *B. burgdorferi* s.s., *A. phagocytophilum*, and Bourbon virus have been detected by PCR in U.S. ALHT collected from vegetation (Thompson et al., 2020; Price et al. 2021, Price et al. 2022, Cumbie et al. 2022, Herb et al., 2023).

In the United States, ALHTs are being collected from new counties and states each year. These changes are reflected in APHIS Veterinary Services Situation Report and ALHT story map.

Status of Occurrence

Identification

Accurate tick identification is important, particularly if the vector-borne disease of interest is absent or not well-established in a region. Molecular testing or a reference laboratory, such as the USDA National Veterinary Services Laboratories (NVSL), in Ames, Iowa, should confirm any taxonomic ALHT identifications from a new State or host (see Appendix A for submission information). State or local laboratories specializing in tick identifications can provide subsequent and additional identifications, if needed.

The National Animal Health Laboratory Network (NAHLN) may be useful to aid in tick-borne pathogen testing. State, university, and public health laboratories may also be able to provide diagnostic capabilities.

Occurrence

The ALHT is established in multiple states and counties within the United States. The "established" designation indicates the species can survive and thrive locally. A minimal observational period of one year is necessary to determine if this tick species can survive and thrive throughout the various local seasonal changes. The critical environmental factors involved in establishment of ALHT across the United States are climate, habitat suitability, and host availability. The ALHT is designated as "reported" (see Table 1 below), at least temporarily, in other states and counties in the United States (Dennis et al., 1998, Ogden et al. 2008, Heath 2016).

Table 1. Occurrence Classes

Occurrence Classes	Description
Established	At least six individual ticks, or at least two of the three host-seeking life stages have been identified in a single collection period (1 year)
Reported	A single life stage (host or vegetation association) is present with no consistent collections over time and space
Absent	No life stage collected in the area with environmental sampling and/or collection of associated small and larger mammalian hosts based on the density of mammalian hosts in the area
No Data	No tick collections have been completed in the area

Epidemiological Investigations and Other Responses

Passive vs Active Surveillance

Passive and active surveillance can be distinguished in the following ways:

Passive Surveillance

- Passive surveillance involves opportunistic tick collections from vegetation and hosts (Mitcham et al. 2017, Ripoche et al. 2018), often via public submissions of specimens.
- Passive surveillance often does not include a defined sampling protocol.

Active Surveillance

- Active surveillance involves systematically collecting ticks from vegetation via flagging/dragging, CO_2 trapping, or collection from hosts (Mitcham et al. 2017, Ripoche et al 2018).
- Active surveillance benefits from the use of grid units to define surveillance areas, with statistical-based sampling used within each sampling grid. This allows for systematic coverage of the areas. Grid units can easily be combined with spatial units (e.g., geospatial layers) relevant to ecological aspects of tick, livestock, and wildlife species surveyed, or the observed distribution of animal or human disease possibly associated with the tick. Geospatial data layers can be used for mapping.
- Active surveillance can be conducted seasonally to understand temporal abundance fluctuations, host associations at various life stages, and monitor changes in distributional trends and patterns.

In collaboration with State/local partners, VS is employing passive surveillance to monitor changes in the geographic distribution of the ALHT in the United States. The dataset associated with tick collections includes recorded environmental collection locations (GPS coordinates, county, state), host-collection locations, and host identifications. Identification of the ALHT, in association with observed clinical illness in animals, warrants additional diagnostic tests and epidemiologic investigation.

If identification of this tick species is linked to a suspected outbreak of tick-associated disease in animals or humans, state agriculture and public health agencies may employ active surveillance methods to assess the risk and potential impact of ALHT on exotic or endemic disease introduction or spread in the United States. VS collaborations with these agencies include discussions on statistical-based sampling designs to ensure surveillance goals are met, recommendations for appropriate tick collection methods in different ecoregions across the United States, and protocols for data management. VS support is available by contacting a VS field office, and surveillance documents are found on the APHIS VS website.

Information Management

Data Management and Reporting

A useful data management platform should provide data formatted for reporting, mapping, modeling, disease risk assessments, economic assessments, laboratory diagnosis, and easy integration into other systems. The VS Emergency Management Response System (EMRS) will maintain premises details for data collected by VS personnel. Standardization of tick survey data from various universities, States, and other Federal agencies is important for future analyses and assessments. Other data management platforms used by other Federal, State, and public agencies can be integrated with EMRS data for VS assessments.

Data for surveyed sites should include the following (see VS Parasite Submission Form VS 5-38 in Appendix A):

Standards

- Date of collection
- Geospatial coordinates
- State name
- County name
- Name of agency or person conducting survey
- Infested host species examined/trapped at the reference site
- Travel history of host and/or owner
- Method used for tick collections (trapping, flagging, dragging, CO_2)
- Tick life stages (larva, nymph, adult [male, female])
- Estimate of the number of ticks collected/submitted

Recommended

- Habitat description, if ticks are collected from environment or vegetation (e.g., shrubs, trees, pasture)
- Sex and age class of host species

ALHT Surveillance

ALHT routinely occur on livestock, wildlife, companion animals, and humans in parts of the eastern United States. It is also collected from vegetation in the environment. Currently, passive surveillance is the most-used method to determine the geographic distribution of this tick species in the United States. Continuing assessment of changes in the geographic and host ranges of ALHT, and of the disease agents associated with it, is necessary to minimize risks to livestock and humans.

New Detection

An epidemiological investigation may be used to determine details related to the detection and identification of ALHT in a new state, new county, or on a new host species.

Initial detections in a state or county should be confirmed at the NVSL or by molecular methods. During any subsequent investigation, a local USDA Veterinary Medical Officer (VMO) or state/local partner should provide a copy of the VS ALHT pest alert fact sheet to the concerned citizen, landowner, or producer. Additional information, such as ALHT situation reports and story maps can be referenced and found on the APHIS VS website. You can also find these documents on the APHIS website that describe the ecology and tick control options for this tick species. Supporting documents are also on the APHIS VS website or in the appendix to this document.

Please note that, after specimen samples are collected for submission and identification, any remaining large infestation of ticks on domestic animals should be treated immediately and before confirmatory identification. Herds should be gathered completely and treated. Treatment may be completed with any acaricide labeled for killing ticks and should be administered according to label directions (see Tick Control section hereinafter for more information). Herd histories should be acquired about movements of animals in and out of the premises and proper traces should be made. Traced herds should be examined for ticks and treated. Detections of ALHT infestations in new states or regions should be routinely treated to achieve control in those areas.

The VMO or state/local partner should discuss clinical disease signs in livestock associated with potential tick-borne diseases with affected producers and leave their contact information for additional inquiries. The VMO or state/local partner should enter investigation data into EMRS after confirmation of ALHT identity. Additionally, the VMO or state/local partner should provide notifications to VS commodity specialists (i.e., equine, cattle, sheep/goats, and/or swine, poultry) after confirming ALHT identification.

If an unusual presentation of clinical signs consistent with an FAD is observed, then the VMO or State/local partner should follow FAD investigation protocols.

Existing and Multiple Detections

After initial confirmation of ALHT presence in a new area, VMOs should work with state agricultural and public health partners to provide educational materials to local producers and the public. This will increase awareness and encourage reporting of any additional tick detections in the area. EMRS is the database for entry of all VS-related tick data.

Scratching for Ticks

Infestations by ALHT on livestock are often heavy and may be perceived by simple visual inspection. However, lesser, or incipient infestations, particularly on cattle, may require detection by human handlers using more specialized manual means (i.e., so-called “tick scratches”).

Inspectors should restrain subject animals safely (e.g., using a chute or alley) and proceed to feel for any attached ticks with their hands. The process generally requires experienced personnel and begins with an examination of the head; between, inside, and outside of the ears; the jawline; frontal body region (brisket); along the flanks; the inguinal area and axillary regions; and under the tail. For cattle, most of the ticks are in the brisket areas and udder regions. Also, human handlers should check areas near the insertion sites of ear tags. This process may also be helpful when looking for ticks on wildlife.

Tick Control

Integrated Tick Control

Due to the complexities of tick ecology and transmission of tick-borne diseases, tick control has many limitations (Torres 2015). In general, single intervention tick-control strategies are only as effective as the duration of the method used (Stafford et al. 2017). For example, the regrowth of modified vegetation will quickly reestablish suitable habitats for ticks, and acaricides generally have reduced efficacies as time passes or as climate changes (e.g., dilution from rainfall). Therefore, multiple tick-control strategies, that are based on tick ecology and are a part of an integrated pest management plan, are important to consider when attempting to increase the chances of reducing tick populations and tick-host contact.

Common tick control methods in agricultural settings include:

- Habitat modifications including pasture vacation
- Acaricide treatment of hosts, vegetation, and equipment

Habitat modifications are often used as part of an integrated pest management plan for tick control and may include mowing grasses, removal of trees, reduction in shade by thinning trees, understory removal, and placement of mulch and surface barriers (Stafford et al. 2017, Piesman and Eisen 2008, Eisen and Dolan 2016). Heath (2016) recommended pasture management by keeping grasses short and treating animals on the pasture to reduce *H. longicornis* populations. Acaricide treatments of host animals alone may not be effective, given that the ALHT spends most of its time in the environment and not on a host. The main goal is to modify the landscape, so ticks are unable to survive there, and/or hosts are not attracted to the area. Pasture vacation is the movement of livestock off an infested landscape to “starve” the remaining ticks. ALHT have wide host feeding preferences and this is not a feasible control solution in areas where rodents, birds, or other wildlife may enter the infested pasture area. Attention to tick infested carcasses is also important, as leaving these on the landscape or moving these to other areas may cause ALHT to reinfest or infest new areas (See Carcass Disposal).

Acaricides and Repellants

To date, acaricide resistance has not been detected in U.S. ALHT populations. When treating for ALHT tick-control products approved by the Environmental Protection Agency (EPA) and the

Food and Drug Administration (FDA) are effective against ALHT although many labels do not currently claim that their products are for the control of ALHT specifically. Laboratory studies indicate that permethrins, phosmet, diflubenzuron, and lambda-cyhalothrin effectively killed exposed ALHTs (Butler et al. 2021). Lee et al. (2015) indicated that organophosphates, amidines, and synthetic pyrethroids may be effective in controlling this tick species in the field over a limited time. The application of amitraz to cattle and permethrin/fipronil to dogs was effective in reducing tick numbers on these hosts (Heath 1980, Duscher et al. 2013). Chemical applications on vegetation should focus on causing mortality earlier in the seasonal questing activity of ticks. They are generally effective in reducing tick densities and tick-host contacts. The seasonal timing of acaricide applications and habitat location are important to have the greatest impact on the various tick developmental stages, on and off hosts. Vegetation in ecotone habitats between mowed recreational areas and forested wetlands in New Jersey were treated with lambda-cyhalothrin spray or granular bifenthrin (both pyrethroids), and these materials successfully controlled ALHT populations, particularly if applied multiple times and sequentially (Bickerton et al. 2021, Bickerton et al., 2022. Bickerton et al., 2023). Bifenthrin 2E®, Paradigm VC®, (pyrethroids), and Sevin SL®, (carbaryl insecticide) are approved for pasture applications. Foster et al.2020 evaluated multiple skin-applied tick repellants against nymphal ALHT in petri dishes, and they found that all licensed tick treatments will work against this tick species, including Coleman SkinSmart®, OFF! Botanicals®, OFF! Deep Woods Sportsmen®, REPEL Plant-based Lemon Eucalyptus®, Sawyer Premium®, and Bite Blocker BioUD®. Repellants and acaricides play a large part in worker safety and the biosecurity practices outlined below.

Biosecurity

When working on ALHT infested and presumed infested areas, it is important to ensure that ticks and pathogens are not spread to subsequent areas and that worker safety is not compromised. The primary purpose of personal protective equipment (PPE) is to minimize exposure to hazards that cause injury and illness, but PPE can also play a vital role in maintaining biosecurity. Biosecurity measures are designed to protect a property from the entry and spread of pests and diseases. To reduce redundancy, protocols to reduce the risk of transferring ticks and pathogens will be included with protocols for personal safety.

Clothing and Footwear

VS recommends the following clothing and footwear to be worn when conducting fieldwork related to ALHT to prevent potential spread of the tick and for worker safety:

- Wear white or light-colored coveralls. The light colors allow for better visibility to identify ticks on your person. White Tyveks are preferred, as these can be treated with permethrin prior to use and before disposal. They are also easy to change between premises. Used Tyveks should be bagged and sprayed with acaricide within the bag before leaving a premises.

- Coveralls should be tucked inside your boots and tape applied around this potential point of entry. Tape can also be applied around wrists to prevent ticks from being able to go up sleeves.
- Spray permethrin-containing products on outer clothing, including boots. before arriving in the field. The permethrin treated clothing should be dry before use. Do not apply in the vicinity of sampling equipment. After applying permethrin, clean the palms of hands (e.g., with soap/water or alcohol swabs) before handling any sampling equipment. Permethrin should not be used directly on skin. It is important to follow the directions on the product label.
- If cloth coveralls are used, they are to be bagged before placing in a vehicle. Cloth coveralls are not to be worn again until washed in hot water and dried on high heat.
- Avoid wearing the same clothes on consecutive days without washing them first to remove ticks. Wash clothes immediately after returning home.

Use Tick Repellant

VS recommends the following when using tick repellent during ALHT fieldwork:

- Apply tick repellants to exposed skin and clothing (See Acaricides and Repellants, page 10). Always read the label before using.
- If skin becomes wet from sweat or water, towel off and reapply the repellent to dry skin.

Perform Tick Checks

- Occasionally during work, and after finishing work at one site and before removing coveralls, conduct a full check of your outer clothing. It is best to work in pairs if possible. Remove any ticks found prior to removing outer clothing and footwear.
- After removing and storing/disposing of outer clothing conduct a body check before leaving the site. Especially examine behind the ears, back of the neck, around the waist, and in and along the hairline. Remove all ticks found.
- Shower as soon as possible to potentially wash away unattached ticks and find attached ticks. Continue to check yourself for a few days after tick surveillance is concluded.
- Remove attached ticks by using fine-tipped tweezers if available. Do not twist the tick as you remove it – this may cause the tick's mouthparts to remain in the skin, increasing the risk of infection.
- Wash the affected area with soap and water and disinfect the bite site.
- Bite sites should be monitored. If flu-like symptoms or a rash develops, seek medical attention.
- A tick bite should be reported to your supervisor.

PPE, Equipment and Vehicles

VS recommends the following PPE protocols for preventing spread of ALHT:

- Check all equipment used at site for evidence of ticks. This should include flagging equipment, clipboards, totes, etc. Equipment not associated with collections may be wiped down with an insecticide.
- Sweep and drag cloths (flags) should either be one time use or examined carefully for evidence of ticks, double bagged and washed in hot water and dried on high heat before using at another site. Flags should NOT be washed with permethrin treated clothing.
- Vehicles should be parked away from grassy areas if possible.
- Vehicle doors, windows, hatch, tailgates, etc. should be left closed unless in use.
- Inspect vehicles, bush hogs, generators, or any other equipment that may have generated CO_2 or warmth while on your site. There have been reports of infestations of ALHT on these types of equipment. Spray with an acaricide before leaving the area.
- Do not enter the vehicle after work is completed unless all outer clothing is removed, properly bagged and thorough tick checks have been completed.

Prevention of the Spread of Veterinary Pathogens

Personnel are expected to follow standard biosecurity procedures when entering and exiting an animal premises.

- Freshly laundered coveralls must be worn. Tyveks are preferred.
- Footwear is to be cleaned and disinfected prior to entering and exiting a premises.
- All equipment used should be disinfected prior to leaving the premises. This includes the handles of the flagging equipment.
- The cloths used for flagging or dragging must not be used on two different farms unless they have been washed in-between. One time use cloths are preferred.
- If there is evidence of a disease on a premises, personnel should not visit another until they have showered and changed clothing.

Carcass Disposal

Early experiences with ALHT infestations on livestock in the United States show that infestations may cause notable mortality rates in host animals, either through massive blood losses or fatal consequences from vectored infections with pathogenic agents. Numerous fatalities in a cattle herd can quickly lead to a problem with carcass disposal. Available options include incineration, burial, landfilling, composting, or chemical digestion. Operators should check with State and local officials for laws and regulations associated with carcass disposal. There may be regulations associated with suspected zoonotic diseases, number of carcasses disposed, ground water regulations, and other conditions that need to be met prior to disposal.

Dead animals with heavy tick infestations may be sprayed with acaricides before disposal; however, caution is necessary to prevent possible acaricide interference with diagnostic tests or disposal methods. Acaricide use should be based on manufacturer specifications.

Incineration may be a good option if carcasses can be moved without dispersing ticks. Ticks may detach and disperse from a dead host as the body cools and blood flow ceases; however, this outcome may depend on tick stages present and their state of engorgement (Piesman 1991, Wang et al. 2001, Tahir et al. 2020). Additionally, partially fed ALHTs have been found questing in the environment (Price et al. 2022), making reattachment of detached ticks a possible threat to other animals. Prompt attention to and treatment of infested carcasses decreases the percentage of ticks leaving the carcass back into the environment.

Composting might be an option, depending on the level of tick infestations, use of acaricides, and suspected presence of zoonotic disease. Acaricide animal treatments may affect composting. Landfill disposal and burying may be options, depending on the level of tick infestations, State/local restrictions on the number of animals that can be disposed in a landfill, or burial requirements associated with regional geology and hydrogeology.

See the following additional references on animal carcass disposal:

- U.S. Environmental Protection Agency Agriculture and Carcass Disposal Guidelines
- California Department of Fish and Wildlife Protocols for Safe Handling and Disposal of Carcasses
- USDA Emergency Carcass Management Desk Reference Guide

Communication/Outreach

Public education will play a major role in the surveillance effort, along with increasing awareness about the ALHT and the potential diseases it may transmit. Materials can be provided to practicing veterinarians, producers, and pet owners through press releases, visits to veterinary clinics, farm visits, information booths at meetings, etc. Fact sheets, distribution maps, story map, tick collection method descriptions, VS Parasite Submission Form (Appendix A, page 18, VS entomologist contact information, and situation reports on the ALHT will be provided on the APHIS VS website.

Training in CO_2 trapping, flagging, and tick collection/shipping is being developed and offered to VS and state field personnel.

As researchers determine more about the ecology of ALHT in the United States, more tools for their control will be developed. State and local animal health authorities should work within the current knowledge of successful control strategies for this tick in the United States to build an integrated control plan for their specific situations. VS entomologists are available for consultation and advice on this process.

References

Bickerton M, McSorley K, and Toledo A. 2021. A life stage-targeted acaricide application approach for the control of *Haemaphysalis longicornis*. Ticks Tick Borne Dis. 12:101581.

Bickerton M, Rochlin I, González J, McSorley, K and Toledo A. 2022. Field Applications of Granular and Liquid Pyrethroids, Carbaryl, and IGRs to Control the Asian Longhorned Tick (*Haemaphysalis longicornis*) and Impacts on Nontarget Invertebrates. Ticks Tick Borne Dis. 13: 102054.

Bickerton M, Gonzalez J, Egizi A, and Toledo A. 2023. Baseline Susceptibility of *Haemaphysalis longicornis* to Organophosphate, Carbamate, and Pyrethroid Acaricides. 2023. Pest Manag Sci. Jun 27. doi: 10.1002/ps.7631. Online ahead of print.
PMID: 37366176.

Breuner N, Ford S, Hojgaard A, Osikowicz L, Parise C, Rosales Rizzo M, Bai Y, Levin M, Eisen R, and Eisen L. 2020. Failure of the Asian longhorned tick, *Haemaphysalis longicornis*, to serve as an experimental vector of the Lyme disease spirochete, *Borrelia burgdorferi* sensu stricto. Ticks Tick Borne Dis. 11: 1–6.

Butler R, Chandler J, Vail K, Holdeman C, and Trout Fyxell R. 2021. Spray and Pour-on Acaricides Killed Tennessee (United States) Field Collected *Haemaphysalis longicornis* Nymphs (Acari: Ixodidae) in Laboratory Bioassays. J Med Entomol. 58(6): 2514-2518.

Cumbie A, Whitlow A, Arneson A, Du Z, and Eastwood G. 2022. The Distribution, Seasonal Abundance, and Environmental Factors Contributing to the Presence of the Asian Longhorned Tick (*Haemaphysalis longicornis*, Acari:Ixodidae) in Central Appalachian Virginia. J.Med.Entomol. 59 (4): 1443-1450.

Dennis D, Nekomoto T, Victor J, Paul W, and Piesman J. 1998. Reported distribution of *Ixodes scapularis* and *Ixodes pacificus* (Acari: Ixodidae) in the United States. J. Med. Entomol. 35: 629-38.

Dinkel D, Herndon R, Noh, S, Lahmers K, Todd S, Ueti M, Scoles G, Mason K, and Fry L. 2021. A U.S. isolate of *Theileria orientalis*, Ikeda genotype, is transmitted to cattle by the invasive Asian longhorned tick, *Haemaphysalis longicornis*. Parasit. Vectors. 14: 1–11.

Duscher G, Feiler A, Leschinik M, and Joachim A. 2013. Seasonal and spatial distribution of ixodid tick species feeding on naturally infested dogs from eastern Austria and the influence of acaricides repellents on these parameters. Parasit. Vectors 6: 76-83.

Eisen, R., Eisen, L. and Beard C. 2016. County-Scale Distribution of *Ixodes scapularis* and *Ixodes pacificus* (Acari: Ixodidae) in the Continental United States. J Med Entomol. 53(2): 349–386.

Eisen L and Dolan M. 2016. Evidence for Personal Protective Measures to Reduce Human Contact With Blacklegged Ticks and for Environmentally Based Control Methods to Suppress Host-Seeking Blacklegged Ticks and Reduce Infection with Lyme Disease Spirochetes in Tick Vectors and Rodent Reservoirs. J Med Entomol. 53: 1063–1092.

Egizi A, Bulaga-Seraphin L, Alt E, Bajwa W, Bernick J, Bickerton M, Campbell S, Connally N, Doi K, Falco R, et al. 2020. First glimpse into the origin and spread of the Asian longhorned tick, *Haemaphysalis longicornis*, in the United States. Zoonoses Public Health. 67: 637–650.

Foster E, Fleshman A, Ford S, Levin M, Delorey M, Eisen R, and Eisen L. 2020. Preliminary Evaluation of Human Personal Protective Measures Against the Nymphal Stage of the Asian Longhorned Tick (Acari: Ixodidae). J Med Entomol.57(4): 1141-1148/

Heath A, Tenquist J, and Bishop D. 1980. Effects of pour-on organophosphate insecticides and a diamidide acaricide on the cattle tick, *Haemaphysalis longicornis*. New Zealand Journal Experimental Agriculture. 8:79-87.

Heath A. 2002. Vector competence of *Haemaphysalis longicornis* with particular reference to blood parasites. Surveillance 29:12-14.

Heath A. 2016. Biology, ecology, and distribution of the tick, *Haemaphysalis longicornis*, Neumann (Acari: Ixodidae) in New Zealand. N Z Vet J. 64: 10-20.

Herb H, Gonzalez J, Ferreira F, and Fonseca D. 2023.Multiple piroplasm parasites (Apicomplexa: Piroplasmida) in northeastern populations of the invasive Asian longhorned tick, *Haemaphysalis longicornis* Neumann (Ixodida: Ixodidae), in the United States. Parasitology. Oct 4:1-29. doi: 10.1017/S0031182023000914. Epub ahead of print. PMID: 37791496.

Lawrence K, Summers S, Heath A, McFadden A, Pulford D, Tact A, and Pomroy W. 2017. Using a rule-based envelope model to predict the expansion of habitat suitability within New Zealand for the tick *Haemaphysalis longicornis* with future projections based on two climate change scenarios. Vet. Parasitol. 243:226-234.

Lee D, Chang K, M. Kim M, Ahn Y, Jo H, and Kim S. 2015. Acaricidal activity of commercialized insecticides against *Haemaphysalis longicornis* (Acari: Ixodidae) nymphs. J. Asia. Pac. Entomol. 18: 715–718.

Levin M, Stanley H, Hartzer K, and Snellgrove A. 2021. Incompetence of the Asian longhorned tick (Acari: Ixodidae) in transmitting the agent of human granulocytic anaplasmosis in the United States. J Med Entomol. 58: 1419–1423.

Magori K. 2018. Preliminary prediction of the potential distribution and consequences of *Haemaphysalis longicornis* using a simple rule-based climate envelope model. bioRxiv https://doi.org/10.1101/389940

Mitcham J, Barrett A, Gruntmeir J, Holland T, Martin J, Johnson J, Little S, and Noden B. 2017. Active surveillance to update county scale distribution of four tick species of medical and veterinary importance in Oklahoma. J.Vector Ecol. 42 (1): 60-73.

Namgyal J, Couloigner I, Lysyk T, Dergousoff S, and Cork S. 2020. Comparison of Habitat Suitability Models for *Haemphysalis longicornis* Neumann in North America to Determine Its Potential Geographic Range. Int. J. of Environ. Res. and Public Health 17:8285. doi:10.3390/ijerph17218285

Ogden D, Lindsay L, Morshed P, Sockett P, Artsob H. 2008. The rising challenge of Lyme borreliosis in Canada. Canadian Communicable Disease Report 34, 1–19.

Pandey M, Piedmonte N, Vinci VC, Falco R, Daniels TJ, and Clark JA. 2022. First Detection of the Invasive Asian Longhorned Tick (Acari: Ixodidae) on Migratory Passerines in the Americas. J Med Entomol. 59(6): 2176-2181.

Piesman J. 1991. Experimental Acquistion of the Lyme Disease Spirochete, *Borrelia burgdorferi*, by Larval *Ixodes dammini* (Acari: Ixodidae) During Partial Blood Meals. J Med Entomol. 28(2): 259-262.

Piesman J and Eisen L. 2008. Prevention of Tick-Borne Diseases. Annu. Rev. Entomol. 53:323–43.

Price K, Graham C, Witmier B, Chapman H, Coder B, Boyer C, and Kyle A. 2021. *Borrelia burgdorferi* sensu stricto DNA in field-collected *Haemaphysalis longicornis* ticks, Pennsylvania, United States. Emerging Infect. Dis. 27(2): 608.

Price K, Witmier B, Eckert R, and Boyer C. 2022. Recovery of Partially Engorged *Haemaphysalis longicornis* (Acari: Ixodidae) Ticks from Active Surveillance, J Med Entomol. 59 (5): 1842–1846, https://doi.org/10.1093/jme/tjac099

Price K, Aryes B, Maes S, Witmier B, Chapman H, Coder B, Boyer C, Eisen R, and Nicholson W. 2022. First detection of human pathogenic variant of *Anaplasma phagocytophilum* in field-collected *Haemaphysalis longicornis*, Pennsylvania, USA. Zoonoses Public Health. 69:143-149.

Price K, Khalil N, Witmier B, Coder B, Boyer C, Foster E, Eisen R, and Molaei G. 2023. Evidence of Protozoan and Bacteiral Infections and Co-infections and partial blood feeding in the Invasive tick, *Haemphysalis longicornis* in Pennsylvania. J Parasitol.109(4):265-273. doi: 10.1645/22-122. PMID: 37436911.

Raney W, Perry J, and Hermance M. 2022a. Transovarial Transmission of Heartland Virus by Invasive Asian Longhorned ticks under Laboratory Conditions. Emerging Infect. Dis. 28(3): 726-729.

Raney W, Herslebs E, Langohr I, Stone M, and Hermance M. 2022b. Horizontal and Vertical Transmission of Powassan Virus by the Invasive Asian Longhorned Tick, *Haemaphysalis* *longicornis*, Under Laboratory Conditions. Front. Cell. Infect. Microbiol. 12:923914. doi: 10.3389/fcimb.2022.923914.

Ripoche M, Gasmi S, Adam Poupart A, Koffi J, Lindsay L, Ludwig A, Milord F, Ogden N, Thivierge K, and Leighton P. 2018. Passive Tick Surveillance Provides an Accurate Early Signal of Emerging Lyme Disease Risk and Human Cases in Southern Canada J Med Entomol. 55(4): 1016-1026.

Rochlin I. 2019. Modeling the Asian longhorned tick (Acari: Ixodidae) Suitable Habitat in North America. J Med Entomol. 56(2):384-91.

Stafford K, Williams S, and Molaei G. 2017. Integrated Pest Management in Controlling Ticks and Tick-Associated Diseases. J. Integr. Pest Manag. 8: 1–7.

Stanley H, Ford S, Snellgrove A, Hartzer K, Smith E., Krapiunaya I, and Levin M. 2020. The ability of the invasive Asian longhorned tick *Haemaphysalis longicornis* (Acari: Ixodidae) to acquire and transmit *Rickettsia rickettsii* (Rickettsiales: Rickettsiaceae), the agent of Rocky Mountain spotted fever, under laboratory conditions. J Med Entomol. 57: 1635–1639.

Tahir D, Meyer L, Fourie J, Jongejan F, Mather T, Choumet V, Blagburn B, Straubinger R, and Varloud M. 2020. Interrupted Blood Feeding in Ticks: Causes and Consequences. Microorganisms 8:910, doi:10.3390/microorganisms8060910

Thompson A, White S, Shaw D, Garrett K, Wyckoff S, Doub E, Ruder M, and Yabsley M. 2021. A multi-seasonal study investigating the phenology, host and habitat associations, and pathogens of *Haemaphysalis longicornis* in Virginia, USA. Ticks Tick-borne Dis. 12:101773.

Thompson A, White S, Shaw D, Egizi A, Lahmers K, Ruder M, and Yabsley M. 2020. *Theileria orientalis* Ikeda in host-seeking *Haemaphysalis longicornis* in Virginia, U.S.A. Ticks Tick Borne Dis. 11: 101450.

Tully B and Huntley J. 2020. A *Francisella tularensis* Chitinase Contributes to Bacterial Persistence and Replication in Two Major U.S. Tick Vectors. Pathogens 9(12): 1037. https://doi.org/10.3390/pathogens9121037

Wang H and Nuttall P. 2001. Intra-stadial tick-borne Thogoto virus (Orthomyxoviridae) transmission: Accelerated arbovirus transmission triggered by host death. Parasitology 122: 439–446.

Appendix A: NVSL Submission Instructions and Form

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to, a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0579-0090. The time required to complete this information collection is estimated to average .083 hours per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information.	OMB Approved 0579-0090 Exp. xx/xx/xxxx

UNITED STATES DEPARTMENT OF AGRICULTURE ANIMAL AND PLANT HEALTH INSPECTION SERVICE VETERINARY SERVICES	**PARASITE SUBMISSION FORM**	NATIONAL VETERINARY SERVICES LABORATORIES 1920 Dayton Avenue Ames, IA 50010 Phone: (515) 337-7514

INSTRUCTIONS: Print in pencil or type. Prepare separate form for each species of animal. Form to accompany specimen. Package specimens well to prevent breakage en route.

1. Submitter's Name		2. NVSL Submitter ID	17. Specimen ID	18. Date collected
3. Name of Business			19. County in which collected	20. State in which collected
4. Address			21. Country in which collected	22. Premises ID
5. City	6. State	7. ZIP Code	23. Latitude	24. Longitude
8. Phone (Area Code)	9. Fax		25. Host origin	
10. Email			26. Host Species (cow, horse, sheep, dog, etc.)	
11. Owner's Name			27. Where found on host (head, ear, flank, back, etc.)	
12. City	13. State	14. ZIP Code	28. Animal ID	
15. Collector's Name		16. Port	29. No. Animals in lot	30. No. Animals infested

31. History or additional information

FOR FIELD LABORATORY USE ONLY (*Optional*)

32. Date Received	33. Date Identified	34. Referral Number

35. Tentative Identification

Identified By:

FOR NATIONAL VETERINARY SERVICES LABORATORIES USE ONLY

36. Case Number	37. Sample Number	38. Date Received

39. NVSL Identification

40. NVSL Accession Number

Identified By: Date

VS FORM 5-38
SEPT 2009

GUIDELINES AND EXPLANATORY DETAILS FOR VS FORM 5-38: PARASITE SUBMISSION FORM
USDA, APHIS, VS, NATIONAL VETERINARY SERVICES LABORATORIES, AMES IA

INSTRUCTIONS: Print in pencil or type. Form must accompany specimen. Package specimens to prevent leakage enroute. Despite best efforts, specimen vials may break or leak alcohol in transit; when that happens, accompanying forms that were completed in ink may become illegible.

1-10. Submitter Information: If the submitter has an ID assigned previously by the NVSL, give the name of the submitter and that ID. For new submitters, or if the ID is not known, please also give the name of the business, mailing address, email address, and telephone and FAX numbers, to whom the identification report will be sent. Specify if there is a preferred method of report delivery; email will be used if no preference is stated. If fax or email is not available, test reports can be mailed but this will delay delivery of your results. Repeat submitters are encouraged to be consistent with the submitter contact information that they provide, as the NVSL keeps a master record. If the test report for an individual submission needs to be routed to a non-standard destination, include special instructions in Block 31 (History or additional information).

11-14. Owner Information: Give the name, city, Sate, and ZIP Code of the owner of the host animal.

15. Collector's name: For multiple collectors, additional names may be given in History or additional information (Field 31).

16. Port: Enter the port of entry, if applicable.

17. Specimen ID: The collector may indicate an identifier for a sample, usually denoting samples taken in series (1, 2, 3, etc.). Please also put that number or other identifier on or in the associated sample container. See also instructions for Referral number (Field 34).

18. Date collected: Enter the calendar date on which the sample was collected from the host animal.

19-21. County, State, and country in which (specimens) **collected:** Specify country only for non-USA collection locales.

22-24. Premises ID, Latitude, and Longitude: Give the NAIS Premises ID and/or geographical coordinates (if known). List coordinates as decimal degrees when possible (e.g., N27.087821 and W92.021484).

25. Host origin: Indicate the county, state, and country (if not the USA) of origin of the host animal.

26. Host species (Cow, horse, sheep, dog, etc.): State the host species from which the parasites were collected. Give only one species, be as specific as possible, and avoid generic terms like "equine", "avian", "reptile", "canine", "feline", etc. Use a separate parasite submission form for each host species. If using "deer", list the kind of deer (white-tailed, mule, etc.). For convenience, names used as examples may be encircled or underlined.

27. Where found on host (head, ear, flank, back, etc.): Indicate one or more anatomical sites where the specimen(s) was or were found. Names used as examples may be encircled or underlined.

28. Animal ID: Give ear/back tag numbers, animal name, or other information that identifies the host animal.

29. No. (Number of) **animals in lot:** Give a count or estimate of the number of individual animals in the group from which the collection was made.

30. No. (Number of) **animals infested:** Give a count or estimate of the number of individual animals in the lot that were infested with parasites; this count will be less than or equal to the number in the lot.

31. History or additional information: This field is open-ended, and may be used to include any additional information pertinent to the collection (e.g., locale, history, quarantine status, additional collectors, etc), observations on host animal condition, or additional details. This field also may be used for notes or requests to the NVSL.

Fields 32-35 are for tentative identifications made at a field laboratory.

34. Referral number: Give submitter's unique identification number assigned to the sample (e.g., foreign animal disease [FAD] investigation number).

Fields 36-40 are for NVSL use only