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CHAPTER 1
INTRODUCTION

1.1 Purpose

This Technical Guide (TG) outlines the purpose of the Department of Defense (DOD) Military Tick Identification/Infection Confirmation Kit [MilTICK], previously known as the Human Tick Test Kit Program (HTTKP). This TG also—

- Describes the customer base that is served by MilTICK, including geographic regions served and the types of tick specimens eligible for submission.
- Discusses the procedures for ordering kits, submitting ticks for identification and testing, and best practices for participating in MilTICK.
- Defines the best practices used within MilTICK to test ticks for human pathogens.
- Discusses the use and reporting of data generated by MilTICK, including the use of MilTICK data to enhance tick-borne disease (TBD) surveillance throughout the DOD.
- Describes other activities conducted by the U.S. Army Public Health Center (APHC) Tick-Borne Disease Laboratory (TBDL) in support of the DOD Lyme Disease Program.

This guide provides an introduction to the use of MilTICK data as actionable evidence on TBD risk for individual tick bite victims, and for supporting vector-borne disease surveillance in military-related populations and environments. It was written with guidance for physicians, physician assistants, nurses, other medical clinic staff, public health professionals (including laboratorians and entomologists), environmental health professionals, and tick bite victims.

1.2 Authority

Army Regulation (AR) 40-5 (2007), Preventive Medicine, para 2-19 and Department of the Army, Pamphlet (DA PAM) 40-11 (2005 (rapid action revision issue date 19 October 2009)), Preventive Medicine, para 4-7 are the authority of this guide. The Director, APHC will provide the DOD Lyme Disease Program and other tick-borne disease prevention programs. The DA PAM will provide pest and vector prevention and control.

The Director of APHC monitors, evaluates, and provides guidance on medical aspects of pest-related injuries and diseases, occupational health exposures from pest control operations, and effective risk communication planning. Procedures used to accomplish these medical functions include lead agent activities for the DOD Tick-borne Disease Program. (Note: With the reorganization of the U.S. Army Medical Command in 2015, APHC is no longer a command, but duties and responsibilities were transferred to the APHC Director. AR 40-5 and DA PAM 40-11 are currently under revision.)

1.3 Abbreviations and Acronyms

A listing of abbreviations and acronyms used in this guide are defined in the glossary.

1.4 Introduction to MilTICK

To help fulfill the medical authority’s responsibilities outlined in AR 40-5 and DA PAM 40-11, MilTICK provides a free tick identification and testing service that offers analysis of tick-borne pathogens of public health significance for military health clinics, military health care providers,
and DOD-affiliated personnel. MilTICK is overseen by the TBDL at the APHC located at Aberdeen Proving Ground, Maryland. MilTICK helps combat the threat of tick-borne diseases to DOD personnel and serves as a “first alert” for tick bite patients and their health care providers.

TBDs, including Lyme disease, are the most commonly reported vector-borne diseases in the United States, and incidences are increasing annually. Tick testing services, such as MilTICK, analyze ticks that have bitten people to determine the health risks associated with that tick bite. Each tick bite has unique risks depending on the species, engorgement, geographic origin, and infection status of the tick. MilTICK assesses each tick individually and reports results back to the tick bite victim or the military treatment facility (MTF) that submitted the tick; they provide actionable evidence that can be used by the bite victim’s health care team in determining the best course of treatment. The data generated by MilTICK are also used in a surveillance capacity to assess the risk of TBD at specific military installations and monitor changes in regional TBD risk.
CHAPTER 2
TICKS AND TICK-BORNE DISEASES

2.1 General Information

Within the United States, ticks transmit a greater variety of pathogens and cause more cases of vector-borne disease (VBD) than any other arthropods, including mosquitoes. Hard ticks (members of the family Ixodidae) are vectors of bacteria, protozoans, and viruses, including many species pathogenic to humans. Incidence of TBD is on the rise across North America, and approximately 95% of VBD incidences reported to the Centers for Disease Control and Prevention (CDC) each year are vectored by ticks. Approximately half of the TBDs known to occur in the northern hemisphere were discovered in the last 25 years.

The ticks of medical importance in the U.S. take a blood meal from a vertebrate host in each active life cycle stage (larva, nymph, and adult). Not all tick species vector the same pathogens; each species is associated with a particular suite of diseases. Different pathogens can coexist in ticks at the same time, so a single tick bite can put the patient at risk for multiple diseases. Different regions of the U.S. harbor different tick species and tick-borne pathogens. Climate modeling suggests that the total area of habitat suitable for ticks is expanding, meaning that ticks and their pathogens are now being encountered in places they did not formerly occupy.

In general, the longer a tick is attached to its host, the more likely it is to transmit pathogens. For this reason, frequent and regular tick checks are recommended, especially after time is spent outdoors in tick habitat.Ticks are small-bodied, particularly in the earlier life cycle stages, making them difficult to spot. A full-body tick check can be conducted with the aid of a mirror or partner. Ticks should be promptly removed to reduce the likelihood of pathogen exposure.

2.2 Tick Species and Their Associated Health Risks

In the subsections below, listed alphabetically by genus, are brief reviews of common human-biting ticks in the United States that MiTICK routinely receives. Included in the review are short descriptions of the tick, their habitat and distribution, and health risks associated with the bite of the tick. Photos, range maps, and references for each tick species can be found in Appendices A and B. All ticks submitted to MiTICK are identified morphologically using taxonomic keys.

2.2.1 Amblyomma americanum, the lone star tick (Appendix A, Figure A-1; Appendix B, Figure B-1).

This is the most common and widespread tick in the southeastern United States, and is an aggressive human biter at all active life stages. Its range extends north through the Mid-Atlantic and has undergone recent expansions. The lone star tick can be encountered in many habitats, including grass, woodland, and transitional areas. Adults and nymphs can generally be encountered throughout the spring and summer months, and clusters of hundreds of tiny larvae can be encountered in the late summer and fall. All life stages bite humans.

The lone star tick is a vector of the bacterial agents of several types of human ehrlichiosis, including human monocytic ehrlichiosis (HME), Ewingii ehrlichiosis, and Panola Mountain Ehrlichiosis. This tick may also infrequently vector the agents of Rocky Mountain spotted fever (RMSF), Tidewater spotted fever (also called Rickettsia parkeri rickettiosis), tularemia, Bourbon virus, and Heartland virus. Lone star tick bites also cause southern tick-associated rash illness
[STARI], which is similar to Lyme disease, but for which the etiologic agent has not been described. The bite of the lone star tick has also been associated with allergic reactions to red meat and dairy products.

2.2.2 *Amblyomma maculatum*, the Gulf Coast tick (Appendix A, Figure A-2; Appendix B, Figure B-2).

This tick is more tolerant of arid conditions compared to the lone star tick. It is distributed throughout the southeastern United States, from Texas in the west to Delaware in the northern part of its range. Historically restricted to coastal areas, it has more recently become established in noncoastal states. Inland populations are more active in the spring to summer, whereas coastal populations are more active in fall to spring. It is primarily associated with grasslands, prairies, and scrub habitat. Generally, only the adults of this species bite humans.

The Gulf Coast tick is a vector of *Rickettsia parkeri*, which causes Tidewater spotted fever, a mild form of rickettsiosis.

2.2.3 *Dermacentor andersoni*, the Rocky Mountain wood tick (Appendix A, Figure A-3; Appendix B, Figure B-3).

This tick can be found in the Rocky Mountains and surrounding areas and in southwestern Canada. It is more tolerant of aridity than the American dog tick, and consequently there is little overlap in their ranges. It is associated with grass and shrub habitat and is typically found at elevations of 4,000 to 10,500 feet. Generally, only the adults of this species bite humans.

The Rocky Mountain wood tick is a vector of the bacterial agents of RMSF, tularemia, and the virus that causes Colorado tick fever.

2.2.4 *Dermacentor occidentalis*, the Pacific coast tick (Appendix A, Figure A-4).

As in its name, the Pacific coast tick is found along the Pacific coast, from Baja California in Mexico to Oregon. It is most frequently encountered in woodland and brush habitats. Generally, only the adults of this species bite humans.

The Pacific coast tick is a vector of the bacterial causative agents of RMSF and tularemia, as well as the viral agents that cause Colorado tick fever and Pacific coast tick fever.

2.2.5 *Dermacentor variabilis*, the American dog tick (Appendix A, Figure A-5; Appendix B; Figure B-4).

This tick is primarily found east of the Rocky Mountains and also has a limited distribution on the West Coast. It is frequently encountered in woodlands, grasslands, and forest edge habitat. It is less tolerant of arid environments than the Rocky Mountain wood tick. The risk of being bitten by an American dog tick is highest in the spring and summer. Generally, only the adults of this species bite humans.

The American dog tick is a vector of the bacterial agents of RMSF and tularemia. Less frequently, it can also vector *Rickettsia parkeri* rickettsiosis (Tidewater spotted fever).
2.2.6 *Haemaphysalis leporispalustris*, the rabbit tick (Appendix A, Figure A-6).

The rabbit tick, also known as the grouse tick, is broadly distributed throughout North and South America. It rarely feeds on humans, preferring instead to take blood meals from rabbits, hares, and ground-dwelling birds.

This tick is a vector of the bacterial agents of RMSF, tularemia, and Q-fever.

2.2.7 *Haemaphysalis longicornis*, the Asian longhorned tick (Appendix A, Figure A-7).

This is an invasive species which is native to eastern Asia, but has become established in Australia, New Zealand, several Pacific Islands, and most recently, the eastern United States. It was introduced into these new areas on cattle, horses, and other animals. The Asian longhorned tick is associated with forests and pastures. In the colder, northern parts of its range, including the United States, this species is parthenogenetic (i.e., females can lay viable eggs without mating).

The Asian longhorned tick vectors several pathogens of veterinary interest, such as the causative agents of canine babesiosis and theileriosis in cattle, and can also cause anemia in livestock in heavy infestations. Presently, it has not been associated with human disease in the United States, but in other parts of the world it has been associated with Powassan virus, Severe Fever with Thrombocytopenia Syndrome virus, Russian spring-summer encephalitis, and other diseases.

2.2.8 *Ixodes scapularis*, the blacklegged tick (Appendix A, Figure A-8; Appendix B, Figure B-5).

The blacklegged tick is also commonly referred to as the “deer tick.” It is found in all states east of the Rocky Mountains and has undergone recent range expansions. It has a wide host range, feeding on mammals, birds, and reptiles. Blacklegged ticks may bite at any time of year, although the risk is greatest in spring, summer, and fall. Any life stage of this species can bite humans, but the tiny nymphs and poppy-seed sized adults pose the greatest public health threats.

The blacklegged tick is a vector of several pathogens of medical interest. These include *Borrelia* bacteria, the causative agent of Lyme disease; *Babesia* parasites, the causative agent of babesiosis; *Anaplasma* bacteria, the causative agent of anaplasmosis, and Powassan virus.

2.2.9 *Ixodes pacificus*, the western blacklegged tick (Appendix A, Figure A-9; Appendix B, Figure B-6).

The western blacklegged tick is found in all states west of the Rocky Mountains and in western Canada, in woodland, grassland, and brush habitats. It is most commonly encountered in California.

The western blacklegged tick is a vector of *Borrelia* bacteria, the causative agent of Lyme disease and *Anaplasma phagocytophilum*, the causative agent of anaplasmosis.
2.2.10 *Rhipicephalus sanguineus*, the brown dog tick (Appendix A, Figure A-10; Appendix B, Figure B-7).

This tick has a cosmopolitan distribution and has been collected in all 50 States, as well as Europe, Asia, Africa, South America, and the Caribbean.

The brown dog tick is a vector of *Rickettsia rickettsii*, the causative agent of RMSF. This species is associated with high incidences of RMSF in tribal lands in the southwestern United States. In other parts of its range (Eurasia and Africa), it also vectors *Rickettsia conorii*, the causative agent of tick typhus, also known as boutonneuse fever or Mediterranean spotted fever.

2.3 U.S. Regional Tick-borne Health Risks

As different tick species are found in different geographic areas, the risks of TBD change depending on where personnel are located. Not every TBD patient recollects getting a tick bite. Ticks are small and are easy to miss, and their bites are often painless, which is why tick checks are important. Ticks may be attached for days before being found, so travel history is also an important factor. Appendix B shows range maps for vector ticks, and Appendix C provides links to APHC fact sheets with more information on TBDs. In the subsections below is a list of TBD in order of prevalence in the U.S. population.

2.3.1 Lyme Disease

Lyme disease or Lyme borreliosis, is the most prevalent arthropod-borne disease in the United States. It is caused by infection with spirochetal (spiral-shaped) bacteria of the genus *Borrelia* (most notably *B. burgdorferi*), which are transmitted to humans via the bite of *Ixodes* ticks such as blacklegged and western blacklegged ticks. It is most common in the northeastern and midwestern United States, where the principal vector is *Ixodes scapularis*; however, cases have been reported from all 50 States. In humans, clinical signs and symptoms of Lyme disease typically include fever, aches, malaise, and an erythema migrans or “bull’s-eye” rash or other rashes, which may or may not be at the site of the tick bite. Treatment with antibiotics such as doxycycline and amoxicillin is most effective if begun promptly after onset of symptoms. A minority of untreated or inadequately treated patients can develop more serious symptoms with neurological, joint, and cardiac involvement.

2.3.2 Ehrlichiosis

Ehrlichiosis is a bacterial infection caused by species in the genus *Ehrlichia*. Humans are infected when bitten by infected ticks, usually lone star ticks in the United States. Clinical signs and symptoms include fever, headache, muscle aches, and fatigue. Severity of symptoms varies with different *Ehrlichia* species. HME is associated with *E. chaffeensis* and can be deadly. More serious symptoms can develop in untreated or immunocompromised patients. It is usually treated with antibiotics such as doxycycline and minocycline.

2.3.3 RMSF and other Rickettsioses

RMSF is caused by the bacterial agent *Rickettsia rickettsii*. Clinical signs and symptoms include fever, headache, muscle aches, nausea, and a spotted rash. The skin rash typically appears around the ankles and wrists before spreading to the trunk, palms, and soles of the feet. More serious complications may include pulmonary edema, cerebral edema, myocarditis, renal
failure, and gangrene. It is important not to delay treatment if RMSF is suspected. *Rickettsia parkeri* causes a form of rickettsiosis which is similar to but generally less severe than RMSF, characterized by signs such as fever, headache, muscle ache, rash, and a characteristic eschar (a cigarette burn-like wound) at or near the site of tick bite.

Doxycycline is the drug of choice for treatment of both diseases.

### 2.3.4 Babesiosis

*Babesia microti* is a protozoal parasite that typically infects rodents. Humans can be exposed when bitten by an *Ixodes* tick that previously fed on a *Babesia*-infected mouse or other rodent, or via blood transfusion from an infected individual. It is most prevalent in the northeastern U.S. and upper Midwest. Clinical signs and symptoms include malaise, anorexia, fatigue, chills, sweating, headaches, and nausea. A combination of antimalarial and antibiotic drugs, such as atovaquone and azithromycin, is commonly prescribed for treatment.

### 2.3.5 Anaplasmosis

Anaplasmosis is also known as human anaplasmosis and human granulocytic anaplasmosis and was previously known as human granulocytic ehrlichiosis. The causative agent of anaplasmosis is *Anaplasma phagocytophilum*, a bacterium. Typical clinical signs and symptoms include fever, headache, chills, and muscle aches. Doxycycline is the drug of choice for treatment.

### 2.3.6 Powassan Virus

Powassan virus disease is a rare, but often serious tick-borne disease caused by the Powassan (POW) virus, a *Flavivirus*. The virus causes inflammation of the brain (encephalitis), and can cause severe damage to the central nervous system. POW virus is the only tick-borne encephalitis found in the United States, though many closely-related tick-borne encephalitis viruses are found across Europe and Asia.

Symptoms of Powassan encephalitis appear suddenly and can develop anywhere from 7 to 30 days after a bite from an infected tick. When POW virus attacks the central nervous system, it kills cells and causes the brain to swell. The membranous coverings of the brain and spinal cord (called meninges) can also swell, causing meningitis. This swelling causes a variety of symptoms ranging from mild to severe. Symptoms range from headache, fever, vomiting, confusion, speech difficulty, and memory loss to life-threatening ones including tremors, seizures, difficulty breathing, and paralysis.

### 2.4 Tick-Borne Diseases Abroad

Ticks are not just a concern in the United States. There are human-biting ticks all over the world, and there are different risks associated with tick bites in different countries. Before deploying, Military personnel should carefully consider the health risks associated with ticks endemic to their destination.
2.4.1 Lyme Disease

Lyme disease is not unique to the United States. It is the most common arthropod-borne disease in Europe, where the usual vector is the sheep tick, *Ixodes ricinus*, and also occurs in Asia, where it is vectored by *I. persulcatus*. Whereas most U.S.-acquired Lyme disease cases are caused by the bacteria *Borrelia burgdorferi*, the causative agent in Eurasian cases is more likely to be another *Borrelia* species such as *B. garinii* or *B. afzelii*. Each of these agents has slightly different characteristics: *B. burgdorferi* is more likely to cause arthritis, whereas *B. garinii* is more likely to affect the central nervous system and *B. afzelii* is more likely to cause lymphocytoma (a hardened nodule on the ear, nipple, or scrotum) and acrodermatitis (a progressive rash that gives the skin a wrinkled appearance).

2.4.2 *Rickettsia* and other bacterial diseases

African tick bite fever is caused by infection with *Rickettsia africae* bacteria, transmitted by *Amblyomma hebraeum* and *A. variegatum* ticks in Africa and the Caribbean. Clinical signs and symptoms include fever, headache, muscle aches, and skin rash.

Tick typhus, also known as boutonneuse fever or Mediterranean spotted fever, is caused by infection with *Rickettsia conorii* bacteria. It occurs in southern Europe, India, and parts of Africa, and is vectored by *Rhipicephalus sanguineus* and related ticks. Clinical signs and symptoms include fever, headache, skin rash, and eschar.

2.4.3 Tick-Borne Viruses

Tick-borne encephalitis [TBE] is caused by the TBE virus in the genus *Flavivirus*. It is transmitted through the bite of infected *Ixodes ricinus* and *I. persulcatus* ticks in Europe and Asia. Clinical signs and symptoms include fever, headache, muscle ache, fatigue, and central nervous system involvement, including meningitis, encephalitis, cognitive dysfunction, tremors, and seizures.

Omsk hemorrhagic fever occurs in Siberia, where humans can acquire it from the bite of an infected *Ixodes* tick, from drinking infected milk, or by contact with infected animals. Clinical signs and symptoms include fever, headache, muscle aches, cough, and internal bleeding.

Crimean-Congo hemorrhagic fever is found in Eastern Europe, Africa, and southwestern Asia. The causative agent is a nairovirus in the family Bunyaviridae, typically vectored by *Hyalomma*, *Dermacentor*, and *Rhipicephalus* ticks; *Hyalomma* ticks are considered the most important vector. Humans can become infected via a tick bite or via contact with infected livestock or human patients. Clinical signs and symptoms include fever, chills, headaches, dizziness, sensitivity to light, muscle pain, and hematomas on the skin. The mortality rate can reach more than 30%, and survivors typically suffer a long convalescent period.

2.4.4 Protozoans

*Babesia divergens* is a parasite transmitted by *Ixodes* ticks in Europe that affects both humans and cattle. Clinical signs and symptoms of infection include fever, anemia, anorexia, depression, weakness, and diarrhea.
2.4.5 Tick paralysis

Tick paralysis is not caused by an infectious agent such as a virus or bacterium, but by a neurotoxin present in some ticks’ saliva. Species associated with tick paralysis include *Ixodes holocyclus* in Australia, *Hyalomma truncatum* and *Rhipicephalus simus* in Africa, and *Amblyomma ovale* in South and Central America. Clinical signs and symptoms include weakness, numbness, and a paralysis that gradually spreads, typically starting with the legs before advancing to the arms, trunk, and head. Symptoms begin after the tick has been attached for several days and reverse promptly when the tick is removed, typically leading to a full recovery within 24 hours; however, death from respiratory paralysis may result if the tick is not located and removed.

Testing services are generally aligned to match regional threats. For identification and testing of ticks removed from DOD personnel abroad, the Public Health Command (PHC)-Europe operates a lab in Landstuhl, Germany that has a tick test kit program similar to MilTICK. For more information or to access tick kits, contact: usarmy.landstuhl.medcom-phcr-e.mbx.phcre-ls-hotline@mail.mil or usarmy.landstuhl.medcom-ph-e.list.ehe-esp@mail.mil. PHC-Europe test ticks from personnel in U.S. European Command, U.S. Central Command, and U.S. African Command.

For tick identification and testing services in the U.S. Pacific Command region, contact:

U.S. Army Public Health Command - Pacific
Medical Entomology Department
ATTN: MCHB-RP-BMED
Unit 45006
APO AP 96343-5006
DSN 315-263-8489; COMM 046-407-8489
CHAPTER 3
HOW TO PARTICIPATE IN MILTICK

3.1 Introduction

MilTICK is a continental U.S.-focused free tick identification and testing service that offers analysis of tick-borne pathogens of military public health significance for military health clinics, military health care providers, and DOD-affiliated personnel. Ticks that bite DOD-affiliated personnel are submitted to MilTICK by mail via tick test kits, which are available through MTFs or by requesting them through the TBDL. Ticks are then identified by species and analyzed for a species-specific panel of tick-borne pathogens. The results associated with each tick are reported back to that individual that was bitten or to a representative at the submitting MTF as quickly as possible. This information provides actionable evidence that can be used by the tick bite victim’s health care team in determining the best course of treatment. The data generated by MilTICK are also used in a surveillance capacity to assess the risk of TBD at specific military installations and monitor changes in regional TBD risk.

3.2 Populations Served by MilTICK

MilTICK serves a variety of populations, all of whom fall under the umbrella of “DOD beneficiary.” Participation in MilTICK is not limited to one Service; MilTICK serves individuals associated with the broader DOD community, including the Army, Navy, Air Force, and Marines. The following populations are eligible to participate in MilTICK:

- Active Duty personnel associated with the Army, Navy, Air Force, Marines, and Coast Guard (e.g., Active Duty Soldiers, Sailors, Airmen and women, Marines).
- National guardsmen and women.
- Reservists in all Services.
- Retired individuals from all Services.
- Civilian personnel working for the DOD or any of the Services.
- Contractors whose current contracts for employment are supported by the DOD or any of the Services (examples include: maintenance or groundskeeper personnel, ORISE).
- Dependents: Includes family (spouse, parents, or children) of all above categories.

3.3 Ticks Eligible for Submission to MilTICK

MilTICK will receive ticks that have been found biting a person in any of the categories of DOD-affiliated personnel outlined in section 3.2. The tick bite does NOT have to have been acquired on a military installation or during work hours. Ticks acquired at home or during recreational activities are all eligible for submission. The tick must have been removed from a human, not a pet or other animal, to be eligible for testing. Ticks found crawling on but not biting a person will not be tested, as there is no risk of pathogen transmission. Damaged ticks that have been burned, crushed, or cut may not be identified or tested depending on the severity of the damage.

3.4 Tick Kit Contents and Instructions for Requesting Kits

Appendix D provides copies of all the documents included in a tick test kit. Tick kits include the following items:
A plastic resealable bag.
A plastic screw-cap 7 ml vial.
A placard with contact information for the TBDL.
An envelope pre-addressed with the address of the TBDL (postage is NOT included).
Instructions for submitting the tick.
A LIDS 850 form which MUST be filled out for the tick to be processed.
A U.S. Department of Agriculture (USDA) permit for shipping tick samples within the boundaries of the U.S.

Tick kits may be requested in bulk by MTFs, clinics, or other military units who suspect they will have a large population of individuals possibly exposed to tick bites. Kits can also be requested in small numbers by individuals. To request tick kits, the TBDL will need—

- A point of contact (POC) to report results to, including an email address and phone number.
- A valid address to ship the kits to.
- The number of kits requested.

Tick kits can be requested in several ways:

- Request online – a link to request tick kits can be located at: https://carepoint.health.mil/sites/ENTO/miltick.
- The TBDL central mailbox can be reached at: usarmy.apg.medcom-aphc.mbx.tickcom@mail.mil.
- TBDL staff can be reached at 410-436-5425 or 410-436-5421 (DSN 584-5425).

3.5 How to Submit a Tick Kit

To submit a tick kit, the tick should first be removed from the person they are biting. Ticks can be saved in resealable plastic bags in the freezer until they can be mailed to MilTICK in a tick kit.

Use the following guidance for tick removal:

- Ticks should be removed as quickly as possible. The most effective way to remove a tick is by using a sharp pair of tweezers to grasp the tick as close to the skin as possible, and gently pull the tick backward and away from the flesh.
- Try not to rupture the tick to minimize the risk of infected fluid coming into contact with the tick bite site.
- Do not use nail polish, nail polish remover or other chemicals, or a lighted match to remove a tick or encourage a tick to detach from a bite site.
- Do not purposefully damage the tick—damaged ticks may not be identifiable or testable.
- Put an antibiotic ointment on the bite site or wash it with soap and water.
- Submit ticks to MilTICK as soon as possible after removal to expedite processing.
The following include ways to submit a tick kit to MilTICK:

- Through a central POC at a clinic or other MTF. MTFs that receive a lot of tick kit requests often have a central POC that receives ticks from tick bite victims and mails the kits to MilTICK. This central POC will assign each tick an internal sample number, and send the tick, the de-identified patient information, and the internal sample number to MilTICK. This prevents MilTICK from receiving information that could be considered personally identifiable medical information that is protected by the Health Insurance Portability and Accountability Act of 1996 (HIPAA).
- By an individual tick bite victim. Ticks can be submitted by individuals mailing a single tick kit, or who to mail in ticks on an ad-hoc basis without travelling to an MTF. Ticks can even be mailed in by individuals who do not have access to tick kits, provided they are a military beneficiary as identified in Section 3.2. Individuals who wish to submit a tick should follow the instructions in the decision tree in Appendix D, Figure D-1.

The instructions on how to send a complete tick kit include (all listed forms below available in Appendix D):

- Page 1 of the LIDS 850 form (or equivalent document printed from the Carepoint tick submission portal) should be completely filled out, including:
  - POC name, address, email address, and phone number.
  - Tick bite victim information including age, sex, Service, and Status.
  - Tick bite information including where the tick bite was acquired, whether it was on- or off-post, and when the tick was removed.
  - Any relevant notes, such as if the tick bite victim has already seen a clinician or has been placed on antibiotics.

- Insert a USDA permit from the current year with each package containing one or more ticks.
- Place the tick in the screw-top vial and ensure the cap is completely tightened.
- Place the vial including the tick in the resealable plastic bag and seal it.
- Put the permit, the completed LIDS 850, and the plastic bag containing the tick inside the vial in an envelope.
- Ensure the envelope has the proper postage—kits with improper or insufficient postage may be significantly delayed in reaching the laboratory.
- Mail to the address on the envelope. For faster delivery, use FedEx or overnight delivery.

In the event that an eligible person has a human-biting tick to submit but cannot readily access a MTF or a tick kit, they should follow the instructions listed in the decision tree in Appendix D, Figure D-1.

### 3.6 What to Expect After Submitting a Tick Kit

Once a tick is received by MilTICK laboratory staff, they will identify that tick to species using morphological features, assess the condition of the tick to include engorgement (how long that tick has been feeding), and determine which tests are appropriate to conduct on the tick. Ticks are only tested for the suite of pathogens for which that tick is a competent vector. Below is a list of topics associated with results reporting, with additional details on each topic:
• **Turnaround time (identification).** Tick species identification and engorgement status are typically reported back to the POC on the same day as the tick is received. This information may be communicated verbally (by phone or in a voicemail), by email, or via the CarePoint site: [https://carepoint.health.mil/sites/ENTO/miltick](https://carepoint.health.mil/sites/ENTO/miltick).

• **Turnaround time (testing).** Testing on a tick and reporting of test results to a POC is typically completed within 2 weeks of receiving the tick. This information may be communicated verbally (by phone or in a voicemail), or by email. A final copy of the LIDS 850 with all fields completed and/or a complete final analytical results report will be mailed or emailed to the POC after testing and initial reporting is completed. Results may also be available via the CarePoint site: [https://carepoint.health.mil/sites/ENTO/miltick](https://carepoint.health.mil/sites/ENTO/miltick).

• **Opting out of information.** If a MTF or clinic submits many ticks each year, they may not wish to receive notification of species identification or negative test results for all ticks. Installation POCs can discuss with TBDL staff if they only wish to receive notification of positive test results. TBDL staff can be reached by email: [usarmy.apg.medcom-aphc.mbx.tickcom@mail.mil](mailto:usarmy.apg.medcom-aphc.mbx.tickcom@mail.mil)

• **Information retention.** Information from tick test kits will be de-identified and kept on file for the ongoing Army mission to conduct surveillance of ticks and their associated pathogens and the evolving risks of TBD to Service members and other DOD beneficiaries.
CHAPTER 4
HTTKP TESTING AND QUALITY ASSURANCE PRACTICES

4.1 Accreditation

MilTICK maintains accreditation for biological testing through the American Association for Laboratory Testing (A2LA). The TBDL is accredited according to the recognized International Standard ISO/IEC 17025:2017 “General requirements for the competence of testing and calibration laboratories.” This accreditation demonstrates technical competence for a defined scope and the operation of a laboratory quality management system. The biological scope for MilTICK is constantly evolving as new tick-borne human pathogens are discovered and new tests are implemented, but broadly covers all testing methods consistently used in the TBDL. As new molecular tests are implemented, they are brought into the TBDL’s scope of accreditation. The A2LA conducts an external audit of all methods in the TBDL every 2 years.

4.2 Tick Testing Practices

MilTICK tests ticks for pathogenic organisms including bacteria, viruses, and protozoans. After a tick specimen is identified to species using taxonomic keys based on morphological characteristics, appropriate pathogen tests are selected based on the species of the tick. The tick specimens are homogenized and the nucleic acids (including DNA, RNA, and/or total nucleic acids [TNA]) are isolated from the specimen. The nucleic acid sample is then tested for the appropriate pathogens using polymerase chain reaction (PCR) technology to amplify selected genetic targets of the pathogens. A positive result indicates that the pathogen of interest is present in the tick sample. Before a positive tick is reported back to a MilTICK customer, a sample must test positive in at least two tests, preferably targeting two separate genetic targets. For all MilTICK laboratory protocols, standard operating procedures (SOPs) and good laboratory practice are followed. This includes the use of positive and negative controls in each test as described below:

- **Positive controls.** Positive controls for each target pathogen are included in each test. Positive controls may consist of a plasmid, cultured cells, or a previously positive MilTICK sample for the pathogen in question. If the positive control does not test positive in a test, the test is considered void and will be re-run.

- **Negative controls.** Negative controls, consisting of purified water substituted for sample, are included in each test. If the negative control does not test negative in a test, the test is considered void and will be re-run.

- **Nucleic acid isolation negative controls blanks.** Every nucleic acid isolation includes one “blank,” which serves as a contamination control. The blank from each nucleic acid isolation run is included in each test using samples from that run. If the blank does not test negative, the test is considered void and will be re-run. If the blank is consistently positive, that run of isolations may have been contaminated.

- **Nucleic acid isolation positive controls.** A subset of nucleic acid isolations are tested for the presence of DNA and/or RNA using quantitative methods (such as a Nanodrop, Qubit, or TapeStation). These are tested by using PCR to test for RNA or DNA spiked into the sample during isolation, and/or by achieving expected prevalence of tick-borne pathogens.
4.3 Quality Control and Assurance

In addition to the positive and negative controls conducted on each test as described above, HTTKP testing integrity is assured through a laboratory quality management system. All PCR tests chosen for use on MilTICK samples are pulled from the peer-reviewed literature; any new test implemented is rigorously reviewed for sensitivity and specificity, and internal SOPs are created and followed before a test is considered routine. Procedures related to tick sample identification, data entry and reporting, nucleic acid isolation, and testing are all governed by TBDL methods and SOPs. New SOPs will be added to the A2LA accredited biological scope as they are completed. Internal audits are conducted on all TBDL methods annually by the APHC laboratory quality management group.

4.4 Technician Training Requirements

Before technicians can conduct activities pertaining to HTTKP samples, they undergo trainings on all necessary TBDL methods as mandated by the APHC laboratory quality management group and A2LA. The trainee must successfully follow the SOP for each method, complete an initial demonstration of capability (DOC), and have their training concurred by the TBDL team lead. Continued DOCs on each method are necessary every few years as mandated by the accrediting body and managed by the APHC laboratory quality management group.
CHAPTER 5
HTTKP RESULTS REPORTING AND USE

5.1 Tick Test Result Reporting

The results of all HTTKP testing are reported either to the tick bite victim or to a central POC at the MTF by sending a final analytical report as described above in section 3.6.

Analyses on ticks are performed via quantitative Real-Time PCR or Conventional PCR as described in section 4.1. If a tick is reported as positive for a pathogen, it means the tick tested positive using two separate molecular tests for the agent of a human disease. Ticks that are reported as negative for a human pathogen did not show any evidence of the presence of that pathogen inside the tick.

The engorgement status of a tick depends on how long the tick was attached to the tick bite victim. If a tick was flat, it may have only been attached a few hours or at most a few days. Ticks that are partly or fully engorged have likely been attached for longer than a few days, and there is a greater chance of an engorged tick transmitting a pathogen to the victim. The ticks that pose the greatest health risk to the tick bite victim will have tested positive for a pathogen and were fully or partly engorged. Engorgement status is evaluated and reported back with the tick identification, as described in section 3.6.

5.2 Use of HTTKP Data to Inform Treatment Decisions

The CDC does not currently recommend tick testing services for the following reasons (from https://www.cdc.gov/lyme/removal/index.html):

- “Laboratories that conduct tick testing are not required to have the high standards of quality control used by clinical diagnostic laboratories. Results of tick testing should not be used for treatment decisions.
- Positive results showing that the tick contains a disease-causing organism does not conclude that you have been infected.
- Negative results can lead to false assurance. You may have been unknowingly bitten by a different tick that was infected.
- If you have been infected, you will probably develop symptoms before results of the tick test are available. If you do become ill, you should not wait for tick testing results before beginning appropriate treatment.”

The APHC TBDL mitigates many of these concerns. The APHC TBDL is an A2LA accredited laboratory that conforms to high standards of testing (see Section 4), and endeavors to report all results within a rapid turnaround time to facilitate the use of this information by the tick bite victim and their medical care team. Subject matter expertise in tick-borne diseases, tick testing, and the interpretation of HTTKP results are available to all personnel who may utilize MiITICK to avoid confusion or misinterpretation of the results. The TBDL is committed to being a resource for accurate, timely, and relevant tick information for the DOD population.

The results reported by MiITICK pertain to the testing of only the ticks submitted. The tick bite victim might have been bitten by other ticks not detected, and should remain alert for any symptoms of tick-borne disease occurring within 1 month of the tick bite. The tick bite victim’s entire clinical spectrum should be evaluated to include the infection status and engorgement.
level of the tick involved. If the tick is positive, the potential for infection is increased, but not confirmed. The risk of disease transmission increases with the increasing engorgement level of the infected tick. If symptoms of a tick-borne disease appear, the tick bite victim should see their medical care team for follow-up care. The use of prophylactic medication, such as doxycycline prescribed after a tick bite, should be considered according to the most recent CDC guidelines: https://www.cdc.gov/ticks/tickborne-diseases/tick-bite-prophylaxis.html. Importantly, while recommended for blacklegged tick bites in Lyme disease-endemic areas, prophylactic antibiotics are NOT recommended for rickettsial or ehrlichial pathogens, and are ineffective against other tick-borne diseases (such as babesiosis).

5.3 Use of HTTKP Data as Biosurveillance

In addition to serving as a first alert for tick bite victims and their medical care team as described in section 5.2, the data are compiled by MilTICK and used as a passive surveillance system to monitor changing threats associated with TBDs to military populations. MilTICK data are stored, de-identified, and analyzed for changing trends regarding where TBDs are located and what sub-populations are at greatest risk of encountering an infected tick. Because this data is passively collected (it is submitted ad-hoc by the DOD beneficiary population and not routinely gathered such as during a field collection study), there are limitations associated with this data. However, MilTICK data is a useful means of assessing human-tick encounters, and fills a gap when active surveillance is too costly or not practical to conduct. This type of passive surveillance data is most powerful when combined with other datasets, such as active surveillance and human case data. MilTICK data are valuable to assess the presence and absence of ticks and their associated pathogens from military installations and surrounding geography where military-related Families may reside. MilTICK data can also be used to document changing trends in pathogen prevalence on military installations.

The data associated with the most recent years of MilTICK can be accessed at: https://carepoint.health.mil/sites/ENTO/miltick. This site can be accessed by anyone with a CAC, and MilTICK data can be visualized by state or by installation. This data can be rapidly accessed and analyzed by anyone looking to assess risks at or near their installation, and used to inform data-driven decision making on TBD threats by DOD leaders. Additional archives of MilTICK data are available through the Vectormap data portal: http://vectormap.si.edu/. 
CHAPTER 6
DOD LYME DISEASE PROGRAM AND OTHER EFFORTS

Since 1986, Lyme and other tick-borne disease-related activities have been conducted at APHC. In addition to MilTICK described in detail above, these activities include:

- Surveillance and other operational studies to detect pathogens in ticks;
- Conducting studies to evaluate tick control or prevention techniques for use by the DOD;
- Providing consultations and subject matter expertise to federal partners about threats associated with ticks and TBD;
- Collaborating with federal, state, and local partners to identify and characterize emerging TBD threats;
- The creation and dissemination of installation and operational TBD threat assessments;
- Developing educational materials which promote personal protective measures and/or increase awareness of tick-related risks; and
- Epidemiological investigations of tick-borne diseases in U.S. Army personnel.

In 1991, the Army was named Executive Agent for a DOD Lyme Disease Program and the APHC (formerly named the U.S. Army Environmental Hygiene Agency) was designated as the Army’s Lead Agency for this program. The DOD Lyme Disease program is the Army’s premier vehicle for the above activities focusing on characterizing and mitigating tick-borne disease threats to DOD personnel. The TBDL at APHC is responsible for the management and implementation of both MilTICK and other activities that fall under the broader umbrella of the DOD Lyme Disease Program.

For additional information on the DOD Lyme Disease Program, TBDL staff can be reached at usarmy.apg.medcom-aphc.mbx.tickcom@mail.mil.
APPENDIX A

PHOTOS OF TICKS OF MEDICAL IMPORTANCE IN THE UNITED STATES

All ticks below are shown in order (from left to right) of adult female, adult male, nymph, and larva. Only human-biting tick life stages are shown for each tick. Photographs were all taken by Mr. Graham Snodgrass, APHC.

Figure A-1. *Amblyomma americanum*, the Lone Star Tick. (left to right) Adult female; Adult male; Nymph; Larva

Figure A-2. *Amblyomma maculatum*, the Gulf Coast tick. Adult female (left); Adult male (right)
Figure A-3. *Dermacentor andersoni*, the Rocky Mountain wood tick. Adult female (left); Adult male (right)

Figure A-4. *Dermacentor occidentalis*, the Pacific coast tick. Adult female (left); Adult male (right)
Figure A-5. *Dermacentor variabilis*, the American dog tick. 
Adult female (left); Adult male (right)

Figure A-6. *Haemaphysalis leporispalustris*, the Rabbit Tick 
(left to right) Adult female; Adult male; Nymph
(Images from Maine Medical Center Research Institute, at: 
https://extension.umaine.edu/ticks/maine-ticks/rabbit-tick/)
Figure A-7. *Haemaphysalis longicornis*, the Asian longhorned tick (adult female)

Figure A-8. *Ixodes scapularis*, the Blacklegged Tick
(left to right) Adult female; Adult male; Nymph
Figure A-9. *Ixodes pacificus*, the western blacklegged tick. Larva (top left); Nymph (bottom left); Adult male (middle); Adult female (right) (Images from tickencounter.org)

Figure A-10. *Rhipicephalus sanguineus*, the brown dog tick. Adult female (left); Adult male (right)
APPENDIX B

RANGE MAPS FOR TICKS OF MEDICAL IMPORTANCE IN THE UNITED STATES

All range maps were downloaded from the CDC website, https://www.cdc.gov/ticks/geographic_distribution.html.

Figure B-1. Lone Star Tick (*Amblyomma americanum*) Range
Figure B-2. Gulf Coast Tick (Amblyomma maculatum) Range

Figure B-3. Rocky Mountain Wood Tick (Dermacentor andersoni) Range
Figure B-4. American Dog Tick (*Dermacentor variabilis*) Range

Figure B-5. Blacklegged Tick (*Ixodes scapularis*) Range
Figure B-6. Western Blacklegged Tick (*Ixodes pacificus*) Range

Figure B-7. Brown Dog Tick (*Rhipicephalus sanguineus*) Range
APPENDIX C

APHC TICK AND TICK-BORNE DISEASE FACT SHEETS

Babesiosis:

DOD Insect Repellent System:

How to Check Yourself for Ticks and Remove an Attached Tick:
https://phc.amedd.army.mil/Pages/Library.aspx?page=9&offset=400

HTTKP:

Human ehrlichiosis:

Human granulocytic anaplasmosis:

Lyme disease:

Protect Yourself from Tick-Borne Diseases:

Spotted fever group rickettsioses (including Rocky Mountain spotted fever):

Tick Control around the Home:
APPENDIX D

HTTKP SUBMISSION FORMS

All current APHC forms can be accessed online at:
https://phc.amedd.army.mil/topics/envirohealth/epm/Pages/HumanTickTestKitProgram.aspx

HTTKP Submission Instructions:

LIDS 850

Current year USDA permit:

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Figure D-1. Decision Tree with Information on How to Submit a Tick to MilTICK for Identification and Testing

The individual will be notified by phone or email once the tick has been received and identified. Results of testing will be reported by phone or email within 2 weeks after the tick has been received.
GLOSSARY

A2LA
American Association for Laboratory Accreditation

APHC
U.S. Army Public Health Center

AR
Army regulation

CAC
Common access card

CDC
Centers for Disease Control and Prevention

DA PAM
Department of the Army, Pamphlet

DOC
Demonstration of Capability

DOD
Department of Defense

DNA
Deoxyribonucleic acid

HIPAA
Health Insurance Portability and Accountability Act of 1996

HTTKP
Human Tick Test Kit Program

MTF
Medical Treatment Facility

POC
Point of Contact

POW
Powassan virus

PCR
Polymerase chain reaction

SOP
Standard Operating Procedure
**TBD**  
Tick-Borne Disease

**TBDL**  
Tick-Borne Disease Laboratory

**TG**  
Technical Guide

**TNA**  
Total nucleic acid

**USDA**  
U.S. Department of Agriculture

**VBD**  
Vector-Borne Disease