Bovine Tuberculosis

This course is designed to provide updated information on the major domestic diseases for which Veterinary Services (VS) has program responsibility. It will provide information on surveillance, disease control and eradication for these diseases. It will also give an overview of the duties of a field Veterinary Medical Officer (VMO) as a support worker of VS animal disease programs and how they interact with other units in APHIS.

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1. DISEASE INFORMATION

a. Agent
Tuberculosis (TB) is a chronic, granulomatous disease, caused by three specific types of bacteria from the Mycobacterium group: *Mycobacterium tuberculosis*, *M. avium* and *M. bovis*. These acid-fast, Gram-positive, rod to filamentous shaped bacteria have a very slow growth rate, resulting in the chronic nature of the disease and are contagious in both humans and animals.

*M. bovis*, the agent for bovine TB, has a great host range and can infect all warmblooded vertebrates, including humans. *M. avium* can affect all species of birds, as well as swine, cattle and humans. *M. tuberculosis* primarily affects humans but can also be transmitted to swine, cattle and dogs. Although the great majority of human cases are associated with *M. tuberculosis*, isolates from approximately 1-5% of cases have proved to be *M. bovis*.

*M. bovis* does not multiply outside of the host, but can typically survive for a few days to a week in the environment. It cannot tolerate prolonged exposure to heat, direct sunlight or dry conditions. The organism can survive longer under cold, dark, moist conditions and persist up to 11 months (18-332 days) at temperatures ranging from 54-75°F (12-24 °F). Feces can remain infective for 6 to 8 weeks and stagnant water for 18 days.

*Mycobacteria* can be cultured on appropriate media, where they multiply approximately once every 20 hours. Because of this relatively slow rate of growth, the disease usually takes many months to develop and in some instances, the organism lies dormant for the lifetime of the host without causing progressive disease.

b. Transmission
*Mycobacterium bovis* can be transmitted from animals to humans and vice versa. The most common route of transmission is through aerosolization of infective droplets (i.e., exhalation or coughing), which are inhaled or ingested by a susceptible host. The risk of exposure is greatest in enclosed areas, such as barns. Cattle can also transfer the organism by nose-to-nose contact or by contact with contaminated feed or water containers. The bacteria are also shed in feces, urine and milk. Transmission through consumption of contaminated raw milk is most common for young animals but also possible for humans, however, Mycobacteria are killed by pasteurization. In some countries, cattle have become infected with *M. bovis* through infected wildlife sources such as deer, elk, badgers (Ireland), brush-tailed possums (New Zealand) and water buffalo (Australia). Although unlikely, humans and other species of animals may also contract bovine TB from consumption of raw or undercooked meat from infected animals.

The risk of bovine TB to humans in the U.S. is low, due to implementation of pasteurization methods and the success of the TB eradication program. Individuals working with infected cattle (i.e., cattle producers, veterinarians, slaughterhouse workers) are at the greatest risk. Most documented human cases have occurred from consumption of raw milk or direct contact with infected animals or their carcasses.
c. Clinical signs

*Mycobacterium bovis* can infect any number of internal organs, however, the lung is most common. Epidemiological data shows the lung as the most affected organ (75%) followed by the retropharyngeal and bronchomedistinal lymph nodes (70%), liver (28%), spleen (19%), uterus (10%), and rarely the udder, intestine or ovary (1% each).2,3,5

Bovine tuberculosis often develops very slowly and may take months to years to develop, therefore animals can be asymptomatic despite the presence of internal lesions. Upon entering the body, the organism elicits a cell-mediated immune response (delayed hypersensitivity) that leads to the production of the granulomatous lesion, known as a tubercule. This lesion contains a core of macrophages, surrounded by giant cells.6 Persistence of the lesions give rise to caseated, necrotic tubercles, which can eventually calcify.

Clinical signs are seldom seen until disease has reached an advanced stage. Some infected livestock seem to be in prime condition, showing no evidence of infection, yet they may be found to be so seriously infected during slaughter inspection that their carcasses must be condemned.1

Clinical signs that may be seen in the advanced stages of bovine TB include progressive emaciation, lethargy, weakness, anorexia, low grade-fluctuating fever, bronchopneumonia, moist cough with dyspnea, tachypnea and possibly superficial lymph node enlargement.4,5 Lung sounds include crackles, wheezes, and silent spots occupied by granulomas.4 Enlarged mediastinal lymph nodes may lead to bloat, while enlarged mesenteric nodes may cause transport failure or obstructions. Involvement of retropharyngeal nodes may cause dysphagia, stridor, and salivation.4

d. Epidemiology

Bovine TB is distributed worldwide. In industrialized countries, control and elimination programs, together with milk pasteurization, have drastically reduced the incidence of disease caused by *M. bovis* in both cattle and humans. In developing countries, however, animal TB is widely distributed, control measures are not applied or are applied sporadically and pasteurization is rarely practiced.7

In 1917, the U.S. State-Federal Cooperative Eradication program was initiated. As a result of program successes, the bovine tuberculosis reactor rate in cattle in the U.S. has been reduced from about 5% to currently less than 0.02%.1 As of 2003, all U.S. states are considered free of bovine tuberculosis (“accredited free”) except Michigan, Texas, California and recently New Mexico (added in 2003).

Bovine TB has been found in the wild deer population of northern lower Michigan since 1994. This has increased the difficulty of eradicating the disease from the state. Currently they are testing that area’s cattle, goats, captive cervids and bison herds in an effort to eliminate the disease. In addition, in concert with wildlife agencies and scientists, the USDA has established surveillance programs for the white tailed deer reservoir, and all manner of intervention strategies are being studied. A request from Michigan for a split state status that would elevate much of the state to the modified accredited advanced status was granted in April 1, 2004.

Texas has also had difficulty eradicating bovine TB across the western part of the state, particularly the El Paso area, due to the close proximity of TB infected dairy herds in Juarez, Mexico. This is despite ongoing testing of large dairy herds and removal of positive animals in the state. Currently a ‘buffer’ region is being developed to help reduce the transmission of the
This includes a total buyout and cessation of dairy production on the Texas side of the El Paso milk shed, which is ongoing.

**e. Diagnosis**

A variety of methods can be used to diagnose bovine tuberculosis. These include tuberculin skin tests, post mortem examinations and various laboratory procedures.

An animal is diagnosed as infected with *M. bovis* based on one or more of the following criteria: 1) a positive response to an official tuberculin skin test 2) diagnostic findings from necropsy examination or slaughter inspection by a Federal or State veterinarian 3) histopathologic examination by a veterinary pathologist 4) positive culture or PCR results for *M. bovis* from selected tissues, 5) decision by a designated tuberculosis epidemiologist (DTE).

### 1) Tuberculin skin tests.

All species infected with *Mycobacteria* develop an immune response (delayed hypersensitivity) to the bacteria, which can be detected by a corresponding tuberculin skin test. Tuberculin is a sterile product made by growing specific *Mycobacteria* species, killing them with heat, and properly diluting and preserving the material,1 which is then called Purified Protein Derivative (PPD) tuberculin. A small amount of this PPD tuberculin is then injected intradermally in the animal(s) being tested to determine infection with or exposure to *Mycobacterium*.

About 72 hours after tuberculin is injected into animals affected with TB, a characteristic inflammatory swelling reaction appears at the point of injection. This reaction is considered a suspicious test result, indicating exposure to one type of *Mycobacteria*.1 Since there is cross reactivity among some mycobacterial species, false-positive reactions may occur in 5-7% of cattle, goat and bison populations due to exposure to avian tuberculosis (*M. avium*), Johne's disease (*M. avium*, subspecies *paratuberculosis*), saprophytic *Mycobacterium* species, or other agents such as *Nocardia* species.4 Therefore, further testing is required to determine a more definitive diagnosis.

There are a variety of possible tuberculin skin tests that may be used to help detect and diagnose bovine TB, based on the situation at hand. The tuberculin skin tests can only detect individuals exposed to *Mycobacteria*, but cannot identify those animals which are diseased. Tuberculin skin test options include the caudal fold tuberculin test, cervical tuberculin test, comparative cervical tuberculin test and single cervical tuberculin test (a test used primarily for cervids).

**a) Caudal fold tuberculin (CFT) test.** The CFT test is used as a primary screening test to identify cattle potentially infected with bovine TB that will need further testing.1 The test involves the intradermal injection of bovine tuberculin into either side of the caudal fold or the vulva. A characteristic inflammatory swelling reaction developing at the point of inoculation within 72 hours (± 6 hours) is considered a positive test result. Further diagnostic methods however are necessary to confirm or refute the presence of *Mycobacterium bovis*.

**b) Cervical tuberculin (CT) test.** The CT test helps to determine if an animal has been infected with *M. bovis*. The test involves the intradermal injection of double-strength bovine tuberculin in the mid-cervical region. Any response noted within 72 hours (± 6 hours) is considered positive for bovine tuberculosis. This test can only be used to classify an animal as a reactor or a negative animal.9 This test will have fewer false negatives, but more false positives compared to the CFT.
c) **Comparative cervical tuberculin (CCT) test.** The CCT test serves to differentiate animals infected with *M. bovis* from those sensitized to tuberculin from other *Mycobacteria* (i.e., *M. avium*). The test involves the intradermal injection of bovine and avian tuberculin PPD into different sites on the same side of the neck of the animal. The injection sites are shaved and thickness measured prior to injection. Seventy-two hours after injection, the responses to the two tuberculins are measured and recorded on a scattergram form. Classification of the animal as negative, suspect or reactor is based on observation and the recorded differences in skin-fold thickness measurements (mm) as plotted on the scattergram.

d) **Single cervical tuberculin (SCT) test - [primarily captive cervids].** The SCT is the official tuberculin test for routine use in captive cervids only and is used for herds whose tuberculosis status is unknown. The test involves the intradermal injection of bovine PPD tuberculin in the mid-cervical region. Visual observation and palpation are done 72 hours (± 6 hours) following injection. This test can only be administered by a State, Federal or designated accredited veterinarian.

2) **Post mortem examination.** Post mortem examination of an animal, either at slaughter or necropsy, can also be used to identify animals infected with bovine TB. Tuberculosis lesions may be found in any organ of diseased animals.

Due to the chronic and insidious pattern of development of bovine tuberculosis, diagnosis can be difficult. In early stages of the disease, lesions can be difficult to find, even during post mortem examination. In later stages, the nodules or tubercules become increasingly evident. Lesions are most often found in the lungs and associated lymph nodes, or the lymph nodes of the head, cervical area and thoracic cavity. Lesions may also develop in the liver, spleen, intestinal tract, and a variety of other organs. Tuberculous granulomas are typically encapsulated, containing thick, yellow caseous pus. They may later become calcified. The organs may be riddled with small miliary tubercles, and in the lungs these may coalesce into a suppurative bronchopneumonia.

3) **Diagnostic laboratory procedures.** A variety of diagnostic laboratory procedures are available to confirm or eliminate a diagnosis of bovine TB following suspicious results from the tuberculin skin tests, gamma interferon, or postmortem examination. The primary laboratory for all TB diagnostic purposes is the USDA-APHIS-National Veterinary Services Laboratory (NVSL) in Ames, Iowa.

a) **Histopathology/Microscopic examination.** *Mycobacterium bovis* can be demonstrated microscopically on direct smears from clinical samples or prepared tissue materials. The acid fastness of *M. bovis* is normally demonstrated with the classic Ziehl/Neelsen stain, but a fluorescent acid-fast stain may also be used. This test will only give a presumptive diagnosis of bovine TB. If the histopathology is typical for *M. bovis* and acid-fast organisms are demonstrated, the specimen is classified as “compatible”; in these cases, it is imperative that the VMO act rapidly to affect quarantine and other appropriate measures. In cases without acid fast organisms, but histopathology is typical, the specimen is classified as “suggestive”; in these cases, follow-up is necessary but is perhaps less critical than with compatible cases.

b. **Polymerase chain reaction (PCR).** Upon finding acid-fast bacteria on histopathology, a polymerase chain reaction test is performed for further diagnosis. Confirmatory diagnosis can be obtained in as little as 2-3 days with 93% accuracy.
c. Culture. For a definitive confirmation (gold standard) of *M. bovis*, material from abnormal lymph nodes or organs such as lungs, liver or spleen should be processed for examination and culture. Cultures are incubated for at least 8 weeks. Growth of *M. bovis* generally occurs after 3 to 5 weeks incubation.

d. Gamma-interferon assay. This assay test is based on the release of gamma-interferon from sensitized lymphocytes during a 16-24 hour incubation period with PPD-tuberculin. The quantification of the lymphokine released is determined by an ELISA test. The sensitivity and specificity of this test is comparable to the intradermal tuberculin test. The Center for Veterinary Biologics (CVB) has approved the gamma interferon test as an official supplemental test in the bovine TB eradication program. The gamma interferon test has been incorporated into the national program for use side by side or as a replacement for the comparative cervical tuberculin test (CCT) in animals that are responders on caudal fold tuberculin test. Also, it is approved for use in TB affected herds in parallel with the skin testing to provide the best opportunity to identify as many infected animals as possible.

e. Other diagnostic tests. Research is being conducted on new serological tests (i.e., nitrite detection, ELISA, fluorescent polarization assays) that may aid in the detection of tuberculosis in livestock and wildlife by making it possible to TB test without having to handle the animals more than one time.

f. Prevention and control

*M. bovis* can be inactivated by a solution of common household bleach (1:9 dilution) and other commercial disinfectants (Nolvasan, Virkon, etc.). The organism is also very heat sensitive and destroyed by pasteurization. However, successful disinfection can only be accomplished after adequate cleaning. The presence of organic matter greatly inhibits most methods of disinfection.

In livestock, bovine TB can be controlled within an affected herd through regular testing and slaughter of any animals that test positive, until the entire herd meets all requirements to be released from quarantine (including multiple full-herd negative tests). However, because there is no method available to ensure that bovine TB has been completely eliminated from an affected herd, APHIS recommends depopulation of affected herds.

The U.S. eradication program, initiated in 1917, has reduced the bovine TB rate from 5% to less than 0.02%. Various surveillance methods are used to detect the disease including examinations made at slaughter, monitoring animals tested for exhibition, interstate movement, herd accreditation and periodic testing of dairies according to milk ordinances.

g. Public health consequences

Bovine TB can be transmitted to humans from infected animals through aerosolization, consumption of undercooked or raw contaminated meat or through ingesting contaminated unpasteurized milk. The highest risk groups include individuals working in close contact with infected cattle (i.e., cattle producers, veterinarians, slaughterhouse workers). Through the efforts of the bovine tuberculosis eradication program, advances in sanitation and hygiene, the discovery of effective drugs and the pasteurization of milk, the incidence of human TB caused by *M. bovis* has decreased significantly. The recent resurgence of human TB has mainly been attributable to *Mycobacterium tuberculosis* and *M. avium*.1
2. HISTORY OF THE DISEASE AND CONTROL PROGRAMS

Bovine tuberculosis has affected animal and human health since antiquity. In the early part of the twentieth century, it was considered the most prevalent infectious disease of cattle and swine in the United States, causing more losses among U.S. farm animals than all other infectious diseases combined.\textsuperscript{1,13} During this time period, the high incidence of humans and cattle infected with \textit{M. bovis} led to the development of the U.S. Cooperative State-Federal Tuberculosis Eradication Program, initiated in 1917. The program involved collaboration between the U.S. Department of Agriculture’s (USDA) Animal and Plant Health Inspection Service (APHIS), State animal health agencies, and U.S. livestock producers.\textsuperscript{13}

When the program officially began, it was estimated that 1 out of every 20 cattle (5\%) slaughtered had bovine tuberculosis\textsuperscript{13} and that approximately 20\% of humans with tuberculosis were infected by \textit{M. bovis}. Additionally, the disease had a great economic impact on producers, as well as on the export trade.\textsuperscript{13} Initially, all cattle herds were systematically tested and all reactors were sent to slaughter. Federal and State agencies shared in the payment of indemnities and premises were cleaned and disinfected after infected cattle were removed. As tuberculosis eradication progressed, the numbers of infected cattle decreased. By 1930, only 1.8\% of tested cattle were reactors (a 64\% reduction).\textsuperscript{13} By 1940, there was a 95\% decrease in the number of slaughter cattle with tuberculosis lesions and the U.S as a whole reached the modified accredited designation status.\textsuperscript{13,16}

Due to the decrease in prevalence of bovine TB, area testing became increasingly inefficient as a method to locate diseased animals. In 1959, emphasis switched from area testing to tracing lesioned animals found by meat inspectors at slaughter. Epidemiological investigation located the herd of origin and any other herds that may have been exposed to the affected animal. All implicated herds were tuberculin skin tested. If tuberculosis was diagnosed, depopulation was highly recommended.\textsuperscript{13}

Great progress has been made in the eradication of bovine tuberculosis from the United States. By the 1990’s, only a few cattle herds each year were found to have tuberculosis. However, tuberculosis in free-ranging (wild) cervids (particularly deer and elk), has been a major impediment to the eradication of tuberculosis. In 1990, Canada banned the importation of U.S. cervid species, following diagnosis of bovine TB in an elk exported by the U.S. This loss of cervid trade led to planning and development of a bovine tuberculosis eradication program for captive (farmed) cervid species.\textsuperscript{13} In May 1994, the Tuberculosis Eradication in Cervidae: Uniform Methods and Rules (UM&R) was published. Considerable progress is again being made, with only 4 captive cervid cases diagnosed between 2000 and 2003.\textsuperscript{13} However, the wildlife reservoir, primarily white tailed deer in Michigan, remains a major and unresolved issue as it impacts final control and eradication of bovine TB. This is compounded by the problems associated with developing effective methods for surveillance in wild cervids. The market for venison is not comparable to that for beef; therefore, effective surveillance through post-mortem examination cannot be supported at the same level.

3. CURRENT CONTROL PROGRAM

Regulations for the bovine TB eradication program are listed in the Code of Federal Regulations (9 CFR – Part 50 and 77)\textsuperscript{15} and the Bovine Tuberculosis Eradication: Uniform Methods and Rules (UM&R).\textsuperscript{9} The objective of the national program is to eradicate the disease from the U.S. so that it
no longer poses a threat to livestock, wildlife and public health.\textsuperscript{15} As a result of this program, the reactor rate in cattle has been reduced from about 5\% to currently less than 0.02\%.

\textbf{a. Surveillance methods}

Currently with the very low prevalence rate of bovine TB, the most efficient way of finding the disease is through a nationwide slaughter surveillance program. Surveillance includes active and passive methods, the use of slaughter, sentinel and outbreak surveillance, as well as mandatory surveillance.\textsuperscript{14}

The goals for surveillance are to 1) detect the last cases of bovine TB in domestic ruminants; 2) measure progress and effectiveness of the eradication program; 3) demonstrate low risk or freedom from disease for trading purposes; and 4) rapidly detect bovine tuberculosis in the event that it is introduced to the United States.\textsuperscript{14} State surveillance is best conducted at a rate that allows detection of tuberculosis in the overall population of livestock at a 0.05\% prevalence level with 95\% confidence.\textsuperscript{9}

\textit{1) Mandatory surveillance.} Through the national eradication program for bovine tuberculosis, all suspected tuberculosis cases are required to be reported by producers, veterinarians, slaughter establishments, and diagnostic laboratories.\textsuperscript{14}

\textit{2) Passive routine surveillance.} Passive surveillance information is obtained from the mandatory reporting of suspected TB cases by veterinarians and diagnostic laboratories, or by testing results required for and obtained from shows, exhibitions or changes of ownership.\textsuperscript{14}

\textit{3) Active routine surveillance.} Active surveillance information is obtained from the inspection and collection of tissue samples at slaughter establishments (slaughter surveillance) or from animals being tested for interstate movement. Additionally, testing information is obtained from dairy herds in accordance to the Pasteurized Milk Ordinance (PMO) Testing.\textsuperscript{14} The PMO outlines requirements necessary for milk producers to market dairy products for public consumption.

\textit{4) Outbreak surveillance.} Surveillance methods used during an outbreak involve tracing and testing all animals (cattle or bison herds) associated with the animal showing evidence of TB (suspect or reactor animal).\textsuperscript{14}

\textit{5) Sentinel surveillance.} Individual herds can be designated as accredited (2 consecutive negative CFT for herd) or qualified (1 negative CFT for herd in last year). For herds to gain and maintain accredited or qualified status, annual herd testing is required. Animals from these herds serve as sentinels based on outcomes from this testing.

Currently, two main methods are used to locate bovine tuberculosis-infected herds: 1.) Identification of infected cattle at slaughter (active routine surveillance).\textsuperscript{14} State or Federal meat inspectors check the lymph nodes and organs of cattle for signs of TB.\textsuperscript{1} When suspect lesions are found, the tissues are submitted to APHIS' National Veterinary Services Laboratory (NVSL) in Ames, IA, for confirmation. 2.) Required testing of animals in areas or populations identified as high risk, due to increased or continued higher than expected occurrences of new cases; this includes expanded testing of dairies in Texas, California and New Mexico, and annual testing of all herds in high risk areas of Michigan.
b. Laboratory testing procedures

An affected herd can be designated when any cattle, bison, captive cervid or goat has been identified by a specially trained veterinary diagnostician, based on all available information and factors. Appropriate considerations include results from official tuberculin tests, postmortem examination or laboratory findings such as histopathology, diagnostic bacteriology and PCR analysis. The primary laboratory for all tuberculosis diagnostic purposes is the USDA - APHIS - National Veterinary Services Laboratory (NVSL) in Ames, Iowa.9

The criteria for diagnosis includes 1) isolation of *M. bovis* from a clinical specimen, 2) histopathology of tissue specimen with pathological lesions compatible with bovine TB, and 3) PCR assay detection of *M. bovis* in tissue specimens. [NOTE: Attempts to grow the bacteria in the laboratory from tissue samples may take up to 90 days or longer since the tuberculosis bacteria are so slow growing.]

c. Epidemiological investigation

Following laboratory confirmation of bovine TB, an epidemiological investigation is conducted to confirm the herd of origin and to trace all other cattle potentially in contact with the infected animal (outbreak surveillance).9 Once found, the complete herd and any other potentially exposed cattle are tuberculin tested or depopulated.1 This includes any adjacent and contact herds, as well as possible source herds for the affected herd. If the herd of origin is diagnosed with *M. bovis*, every effort is made to eliminate all animals in that herd. Indemnities are paid to help compensate owners for their losses.1 If the herd cannot be depopulated, it is held under quarantine and tested repeatedly until all evidence of infection is eliminated and all restriction requirements (including repeated testing at defined intervals) are met.1

Veterinary epidemiologists also attempt to determine the date the herd was most likely infected. They then undertake a concerted effort to trace all cattle that moved into or out of the affected herd subsequent to the date of most probable exposure, in an effort to determine the most probable source of the infection and where it may have spread.1

d. Accreditation

1) Area (State) accreditation. The eradication program has instituted designation levels for a state based on their bovine tuberculosis prevalence. A state’s designation level affects the movement requirements of animals into and out of the state. The different status designations possible (from highest to lowest) are Accredited Free, Modified Accredited Advanced, Modified Accredited, Accreditation Preparatory, and Non-accredited.

All States must have the authority to enforce and comply with the provisions of the Uniform Methods and Rules for Bovine Tuberculosis Eradication. Additionally, states must maintain clinical and epidemiologic surveillance of animal species at risk of tuberculosis at a rate that allows detection of tuberculosis in the overall population of livestock at a 0.05% prevalence rate with 95% confidence.15

a) Accredited Free. Cattle or bison that originate in an accredited-free State or zone may be moved interstate without restriction. For a State to be recognized as Accredited Free (desired goal for all states) for bovine TB, the State must have laws and regulations governing livestock dealers. It must also maintain surveillance of cattle in marketing channels and require animal records that allow animal health officials to trace infected animals back to their source.1,9 There must be zero prevalence of affected cattle and bison herds, and no findings of tuberculosis in any cattle or bison
in the State for the previous 5 years. Exceptions to the 5 year rule are made for States that have been Accredited Free previously but subsequently were downgraded in status. These States are eligible to re-attain Accredited Free status 2 years after the last affected herd has been depopulated or released from quarantine.

In the case that an affected herd is detected within an accredited free state, the herd is depopulated and an epidemiologic investigation is completed within 90 days. If there is no evidence of the spread of TB, the State may retain its accredited-free status. However, if two or more affected herds are detected within a 48-month period the State will be reduced in classification to modified accredited advanced status.15

The accredited-free State or zone status must be renewed annually. To qualify for renewal, a State must submit an annual report to APHIS prior to November 30. This report should show the amount of testing and slaughter surveillance that has been conducted. Approval and maintenance of a State’s status requires yearly review and approval by the Administrator of APHIS. The Designated Tuberculosis Epidemiologist (DTE) must review reports of all testing for each zone within the State within 30 days of the testing.15

b. Modified Accredited Advanced. This status is designated to a State that has less than 0.01% prevalence of bovine tuberculosis in the total number of herds of cattle and bison in the most recent 2 years. The Administrator may allow a State or zone with fewer than 30,000 herds to have up to 3 affected herds for the most recent 2 years depending on review of the State resources and situation.14

c. Modified Accredited. This status is designated to a State that has less than 0.1% prevalence of bovine tuberculosis in the total number of herds of cattle and bison in the most recent year. The Administrator may allow a State or zone with fewer than 10,000 herds to have up to 10 affected herds for the most recent year, depending on review of the State resources and situation.14

d. Accreditation Preparatory. If a State has less than 0.5% of the total number of herds of cattle and bison in the State or zone, it is designated as accreditation preparatory. No U.S. States are listed at this status level as of 2004.
e. Nonaccredited. A State that does not meet the standards of the Bovine TB UM&R or has a bovine tuberculosis prevalence at 0.5% or greater is designated as non-accredited. No U.S. States are listed at this status level as of 2004.

Table 1 shows the various area (state) accreditation levels, status requirements and their movement restrictions for the bovine tuberculosis eradication program. The majority of states are Accredited Free. In 2004, Texas, California and New Mexico were Modified Accredited Advanced. Michigan was granted split State status with all or part of 13 counties being Modified Accredited and the majority of the stated designated Modified Accredited Advanced. Currently (2004) no states are Accreditation Preparatory or Non-accredited.

2) Herd Accreditation. Livestock owners can achieve accredited TB status for their individual herds by following the “Accredited Herd Plan” in the Bovine Tuberculosis Eradication, Uniform Methods and Rules.9
**a. Cattle.** For a cattle herd to qualify as accredited, all cattle 24 months of age or older and cattle of any age that are not natural additions to the herd, must attain negative findings on two annual TB tests.\(^1\) Strict record keeping of all animals added or removed is required.\(^9\)

When herds have passed at least two consecutive annual caudal-fold tuberculin tests, have no other evidence of bovine TB, and meet the standards of the UM&R for Bovine TB eradication, they are eligible to be USDA accredited bovine TB-free herds. The benefits of this status are that animals can move interstate freely and from one accredited herd to another, and there is only an annual whole herd test for any animal movement from the herd.\(^9\) Such herds shall be issued a certificate by the local State or Federal animal health officials.\(^9\)

To qualify and continue as an accredited herd, cattle and bison must be tested annually within 9 to 15 months of the anniversary of the original test. Livestock from any herd in an accredited-free State may be added to an accredited herd without a qualifying test.\(^1\)

**b. Goats.** For accreditation or re-accreditation of goat herds, all goats 12 months of age and older shall be tested.\(^9\) New additions to an accredited herd must have a negative test result within 60 days prior to entering the premises or the accredited herd and must be kept in isolation from all members of the accredited herd until they have a negative result on a tuberculosis test administered at least 60 days after date of entry.\(^9\)

**c. Captive cervids.** Testing of captive cervids for individual herd classification must include all captive cervids over 1 year of age and animals 1 year of age and under that are not natural additions.\(^15\) To meet the requirements for accredited herd status, the herd must pass at least three consecutive official tuberculin tests conducted at 9-15 month intervals with no evidence of bovine tuberculosis. Additions to an accredited herd should be isolated from all members of the accredited herd until the addition has had a negative result 90 days prior to entry and at least 90 days following entry into the accredited herd.\(^15\)

### 4. CONTROL PROGRAM STATUS

The bovine TB program has achieved much since its initiation. By 2002, all 50 states plus the U.S. Virgin Islands and Puerto Rico remained at or had at one time achieved accredited-free status for bovine TB in cattle and bison (only a small zone of extreme west Texas has never been recognized as accredited TB free).\(^8\) [Note: In 2003, California and New Mexico were reduced in status to ‘modified accredited advanced’ following the identification of three and two tuberculosis affected herds, respectively].

Currently all U.S. states are considered free of bovine tuberculosis (accredited free), except Michigan, Texas, California and recently New Mexico (2003).

Slaughter surveillance had increased by 62% over FY 2001 and 500% over the 1999 levels. Standard surveillance measures for all species (cattle, bison and cervids) have also been developed.\(^11\) The gamma interferon test was approved by the USDA-Center for Veterinary Biologics (CVB) as an official supplemental test in the bovine TB eradication program. Additionally, an electronic method to gather and report monthly TB test data was developed and implemented.\(^11\)
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<th>Status Level</th>
<th>Movement Requirements: Breeding Animals</th>
<th>Movement Requirements: Steers and Spayed Heifers</th>
<th>Prevalence Level (risk)</th>
<th>Qualification for Advancement to the Next Level</th>
</tr>
</thead>
<tbody>
<tr>
<td>Accredited Free</td>
<td>No test requirement out of State or zone</td>
<td>No test requirement out of State or zone</td>
<td>0% herd prevalence, or one infected herd during the previous 12 months and meets the minimum standards set by the program UM&amp;R.</td>
<td>If one infected herd is detected in a State/zone considered to be Accredited Free, the herd must be depopulated, and a complete epidemiologic investigation of the outbreak must be complete within 6 months from detection of the infected herd or face status reduction.</td>
</tr>
<tr>
<td>Modified Accredited Advanced (NEW STATUS)</td>
<td>One negative TB test required prior to interstate/interzone movement</td>
<td>No test requirement out of State or zone</td>
<td>Less than 0.01% herd prevalence or 1-3 infected herds in States/zones with fewer than 30,000 herds, and meets the minimum standards set by the program UM&amp;R.</td>
<td>Must follow the UM&amp;R and must not have an infected herd for the previous 5 years. If a State/zone has depopulated all affected herds, then the State/zone would qualify for Accredited Free status in 3 years.</td>
</tr>
<tr>
<td>Modified Accredited</td>
<td>Herd of origin TB test requirement and 1 individual animal TB tests required prior to interstate/interzone movement. Cervidae from Qualified &amp; Monitored herds require only 1 negative test. Cattle or cervids from Accredited herds may move if Accreditation test was within 12-months of movement.</td>
<td>One individual animal TB test required prior to interstate/interzone movement.</td>
<td>Less than 0.01% herd prevalence, or a maximum of 5-10 infected herds or a maximum of 10 infected herds in States/zones with fewer than 10,000 herds, and meets the minimum standards set by the program UM&amp;R.</td>
<td>Must follow UM&amp;R AND verify a herd prevalence less than 0.01% or less than 1-3 infected herds in States/zones with less than 30,000 herds for 2 consecutive years.</td>
</tr>
<tr>
<td>Accreditation Preparatory (NEW STATUS)</td>
<td>Herd of origin TB test requirement and 2 individual animal TB tests required prior to interstate movement. Cervidae from Qualified &amp; Monitored herds require only 1 negative test. Cattle or cervids from Accredited herds would require 1 negative test.</td>
<td></td>
<td>Less than 0.05% herd prevalence, and meets the minimum standards set by the program UM&amp;R.</td>
<td>Must follow UM&amp;R AND verify a herd prevalence less than 0.1% or less than 10 infected herds in States/zones with fewer than 10,000 herds, for 1 year.</td>
</tr>
<tr>
<td>Non Accredited</td>
<td>PROHIBITED</td>
<td>PROHIBITED</td>
<td>Unknown or greater than or equal to 0.5%, or doesn’t meet minimum standards set by the program UM&amp;R.</td>
<td>Must have authority to enforce UM&amp;R, maintain a herd census, establish a surveillance system, and have determined a herd prevalence rate of less than 0.5%.</td>
</tr>
</tbody>
</table>
5. **CONTROL PROGRAM AUTHORITY**

State laws and/or regulations should provide the authority to apply a tuberculin test to any animal or herd when deemed necessary by State or Federal officials. Tuberculin tests shall be applied by a veterinarian employed in a full-time capacity by the State or USDA or by an accredited veterinarian.9

A Designated Tuberculosis Epidemiologist (DTE) is a State or Federal epidemiologist specially trained and designated to make decisions concerning the use and interpretation of diagnostic tests for tuberculosis and management of tuberculosis affected herds.14 They have the responsibility to determine the scope of investigation, determine the status of animals and herds, assist in the development of individual herd plans, and coordinate disease surveillance and eradication programs within the geographic area of their responsibility.14

6. **ROLE OF THE VMO IN THE BOVINE TUBERCULOSIS ERADICATION PROGRAM**

**a. Quarantine procedures**

All herds in which suspect or reactor animals are disclosed shall be quarantined. Exposed animals must remain on the premises where disclosed unless a State or Federal permit for movement has been obtained. Movement for immediate slaughter must be directly to a slaughtering establishment where approved State or Federal inspection is administered.9

Herds indicated as the source(s) of slaughter traceback case investigations shall be placed under quarantine within 30 days of notification, and a herd test of all eligible livestock shall be scheduled.9

**b. Acceptable TB testing procedures**

Diagnostic field testing for bovine tuberculosis involves an official tuberculin test which is applied and reported by an approved veterinarian in accordance with the bovine tuberculosis eradication UM&R. Any animal tested with an official tuberculosis test must bear official identification in the form of an official eartag, or other approved identification device or method.14

Official tests for cattle, bison and goats include the cervical tuberculin test (CT), caudal fold tuberculin test (CFT), and the comparative cervical tuberculin test (CCT). The gamma interferon assay is an official supplemental test for cattle only.

The official tuberculin tests for captive cervid herds include the single cervical tuberculin test (SCT) and the comparative cervical tuberculin test (CCT).

1) **Cervical tuberculin (CT) test** is recommended for use with herds known to be affected by bovine tuberculosis. It is required as the initial test for testing exposed cattle or bison from suspect herds. This test can only be administered by a State or Federal regulatory veterinarian.9 All animals with a positive response should be classified as reactors.9 A positive response includes erythema or swelling noted at the injection site 72 hours (±6 hours) from administration of bovine PPD tuberculin.
2) **Caudal fold tuberculin (CFT) test** is the primary diagnostic test in cattle, bison or goat herds when the tuberculosis status of the herd is unknown. This test is administered by a federal or state veterinarian and is routinely performed by Federally accredited private practitioners. This test should only be used as a herd screening test. All positive responses should be recorded and the animal classified as suspect, unless a reactor classification is indicated. This test should be followed by further diagnostic methods (ie, CCT) to determine the presence of *M. bovis*.

3) **Comparative cervical tuberculin (CCT) test** is performed on animals that demonstrate a response to the CFT. It is used to determine if the CFT reaction was more likely due to infection with or exposure to *M. bovis* or *M. avium*. This test can only be administered by a State or Federal regulatory veterinarian.

For cattle and bison, the test must be performed within 10 days or after 60 days of the initial CFT. All cattle on the farm must be quarantined until results from the CCT are obtained. Responses should be recorded and plotted on the CCT scattergram. Classification is determined by which zone the animal is plotted within. If an animal is determined to be a “suspect” on two successive CCT tests, it is then classified as a reactor.

For captive cervids, the CCT test may only be used for retesting captive cervids classified as suspects with the SCT test. The CCT retest must be done within 10 days or after 90 days of the SCT. Injection of the PPD should be on the opposite side of the neck as the SCT was administered. If the CCT test results show the captive cervid to be a suspect, the test is repeated at least 90 days after the first CCT. If an animal is determined to be a “suspect” on two successive CCT tests, it is then classified as a reactor. The CCT test may only be used for retesting captive cervids classified as suspects. It may not be used as a primary test for herds of unknown tuberculosis status.

4) **Single cervical tuberculin (SCT) test** – [primarily captive cervids]. This test is the official tuberculin test for routine use in captive cervids. It should be used as a screening test for herds whose tuberculosis status is unknown. A positive test result is suspect for *M. bovis* infection. Suspect animals, as well as, the herd must be quarantined. Additionally, suspects should be slaughtered or retested by a CCT test. Animals positive on the CCT test should be classified as reactors.

5) **Gamma interferon assay [cattle only]**. This test may be used in cattle herds only when approved by State/Federal Animal Health Officials as an official test for use in the state and with the concurrence of the designated TB epidemiologist (DTE) and the Veterinary Services (VS) regional tuberculosis epidemiologist. It is used in cattle herds as follows: a) in parallel testing with the comparative cervical tuberculin (CCT) test; b) as a replacement for the CCT test for retesting CFT test suspects. (CCT test suspects must be retested negative on the CCT test prior to reclassifying such suspects as negative.); or c) in parallel with the caudal fold tuberculin (CFT) test in affected herds.

Results of the bovine Interferon Gamma Assay shall be interpreted and reported by the laboratory as positive or negative. Animals positive on the bovine Interferon Gamma Assay will be classified as tuberculosis test suspects unless the DTE determines that a reactor classification is justified.
c. Reading the CCT test result
Seventy-two hours (±6 hours) after the administration of the PPD, the two inoculation sites are analyzed for erythema, swelling and skin thickness. This must be performed by the same veterinarian who initially administered the tuberculin. Based on observation, measurements with calipers and comparison of the two injection sites, test results are plotted on an official USDA CCT scattergram plot (Figure 1). The plotted results can then be used to classify the animal as negative, suspect or a reactor. Approved scattergrams are available for interpreting results in cattle, bison and reindeer.

1) Negative. For cattle and bison, an animal is considered TB negative if the CCT test results are within the negative range of the scattergram plot. For captive cervids, an animal is considered TB negative if the response to bovine PPD measures less than 1 mm. If the entire herd tests negative, the quarantine is released.

2) Suspect and identification. If cattle or bison CCT test results are plotted midway on the scattergram, the suspect animal must either 1) be removed from the herd, euthanized and submitted for necropsy and further tests, or 2) the herd remains quarantined an additional 60 days at which time the suspect animal can be retested. Captive cervids are classified as suspect if their bovine PPD is 1-2 mm and equal to or greater than their avian PPD response, OR if their bovine PPD is greater than 2 mm but equal to their response to avian PPD.

To be eligible for Federal indemnity, exposed suspect cattle and bison shall be identified by branding the letter “S” on the left hip near the tailhead and by tagging the animal with an approved metal eartag (bearing a serial number) in either ear of the animal. In lieu of branding, animals may be accompanied to slaughter by a Federal or State representative or be shipped in an officially sealed vehicle.

3) Reactor and identification. If the CCT test results for cattle and bison fall within the reactor range of the scatterplot, the animal is considered a bovine tuberculosis reactor. Captive cervid animals having a response to bovine PPD greater than 2 mm and at least 0.5 mm greater than their avian PPD should be classified as a reactor. Additionally if any animal is determined as a suspect on two successive CCT tests, it should be classified as a bovine tuberculosis reactor.

Once classified as a reactor, an animal becomes subject to federal regulations. Reactor animals shall not be retested or reclassified. The reactor animal must be removed from the farm, euthanized and submitted for diagnostic testing within 15 days. All remaining animals on the farm are quarantined until results are obtained and a federal movement permit is granted.

Reactor cattle and bison should be identified by branding the letter “T” on the left hip near the tailhead and by tagging the left ear with an approved metal eartag bearing a serial number and inscription “U.S. Reactor” or similar State reactor tag. In lieu of branding, the reactor animal(s) should be tattooed with the letters “TB” in the left ear and should also have the letters sprayed on the left hip with yellow paint. The reactor(s) may be shipped to slaughter in an officially sealed vehicle or accompanied to slaughter by a State or Federal regulatory official.
d. Reporting
A report of all approved tuberculin tests shall be submitted in accordance with State and Federal requirements. The report shall include the individual identification of each animal by an official eartag number or tattoo, age, sex, and breed and a record of all responses listing the size of the response and test interpretation.

e. Managing an infected herd - Actions to take
When a reactor animal is confirmed to indeed have bovine TB, a farm owner/producer is notified and one of two options may be followed, either complete herd depopulation or a test and remove option.

Figure 1. Photo image of a tuberculosis scattergram.
1) Depopulation. When a tuberculous animal is identified, producers are encouraged to consider whole herd depopulation. This involves destruction of all livestock exposed to bovine TB in the herd, before any restocking of the premise with cattle, captive cervids, bison or goats. Following depopulation, the farm is thoroughly cleaned and disinfected, and may only be repopulated following approval by a DTE or following one year from removal of the animals. Current policy allows for indemnification payments for the fair market value of animals destroyed, not to exceed $3,000 per animal.9

For newly assembled herds placed on premises where a tuberculous herd was depopulated, two annual herd tests shall be applied to all cattle and/or bison. The first test must be applied about 6 months after assembly of the new herd. If the premises were vacated for over 1 year, these requirements can be waived by the DTE.9

Cattle and bison in feedlots known to be exposed to tuberculous cattle or bison should be quarantined and shipped under permit directly to slaughter. The feedlot or affected portions of the feedlot should be vacated, cleaned and disinfected following the removal of affected and exposed cattle or bison to slaughter.9

2) Test and remove. Following a confirmed case of TB, a farm remains under quarantine until all animals are found negative for TB and stringent testing requirements have been met. Dairy farms may continue to sell milk while under quarantine, if all bovine TB reactors are removed from the farm. Movement of animals for immediate slaughter must be directly to an establishment with State or Federal inspection.

For herds with confirmed M. bovis infection, USDA-APHIS recommends depopulation. If however, this is not done, the herd shall remain under quarantine until the herd passes two tuberculin tests at intervals of at least 60 days (three tuberculin tests at intervals of at least 90 days are required for captive cervid herds), one additional test after 180 days and three annual whole-herd tests.

All animals moved from a quarantined farm shall be shipped directly to slaughter and shall be accompanied by a shipping permit (VS1-27) issued by a State of Federal representative. After the quarantine is released, the herd must undergo two additional whole-herd tests. The first test is to be applied about 1 year after release from quarantine.9 This entire process, of approximately eight whole herd tests, can take up to 48 to 60 months. This option can be costly and have long term implications for farm operation.

f. Movement of animals
No animal with a response to an official tuberculin test is eligible for interstate movement unless the animal is classified as “negative” from M. bovis on an official comparative cervical tuberculin test9 or gamma interferon test.

No animal with a response to any tuberculosis test is eligible for international movement.

g. Premise cleaning and disinfection
Following identification of a tuberculosis reactor animal, all premises must be properly cleaned and disinfected within 15 days after removal of the tuberculosis-affected or -exposed cattle or
bison. Feedlots or portions of feedlots that have contained affected or exposed cattle or bison shall be vacated, cleaned and disinfected following the removal of such animals to slaughter.9

h. Quarantine release
If a suspect animal is found negative for TB following either confirmatory testing, or slaughter and histopathology and culture, the herd may be released from quarantine. If the suspect is classified as a reactor, the herd may only be released from quarantine if no gross lesions are found upon slaughter, histopathology and culture, and the remainder of the herd is found negative on a whole herd test. A herd retest is recommended in 60 days for cattle and bison and 90 days for captive cervids.9

A captive cervid herd determined to be affected by bovine TB must be quarantined until the herd has tested negative on three successive whole herd tests. The first test given must be 90 days or more after the last reactor test. In addition, the herd must be given five consecutive annual whole herd tests after release from quarantine. As an alternative to testing, the herd may be depopulated.

Sale of feeder calves from quarantined herds shall be restricted. Feeder calves under 12 months of age that have passed a CFT test within 60 days may be permitted to move intrastate to a quarantined feedlot.9

i. Surveillance - Traceback
Disclosure of tuberculosis in any herd shall be followed by a complete epidemiologic investigation. Evaluation and testing should be implemented promptly on all adjacent and contact herds as well as possible source herds for the affected herd.9

A tuberculosis infected feedlot shall be handled in the same manner as an affected herd, with epidemiologic investigation of animal movement into and out of the feedlot.9

7. ROLE AND OTHER AGENCIES, STATES AND INDUSTRY

Under the Cooperative State-Federal TB program, USDA and/or States provide personnel to trace and test herds and animals epidemiologically implicated as a result of TB infection identified at slaughter, with all indicated followup. Some States provide indemnity to owners. Public health services require TB tests of dairy herds in states that are not either accredited TB free or modified accredited advanced states.16

8. REFERENCES


