NVAP Reference Guide: Tuberculosis (Control and Eradication)

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Appendix

Bovine tuberculosis (TB) is a contagious, infectious, communicable disease of animals and humans caused by *Mycobacterium bovis*. It is commonly a chronic, debilitating disease but occasionally may assume an acute, rapidly progressive course. TB is a widespread zoonosis of global magnitude and affects nearly all species of vertebrates.

Disease is spread by direct contact, inhalation of droplets expelled from infected lungs, and ingestion of contaminated feed or milk. All accredited veterinarians must immediately report every suspected or diagnosed bovine TB case promptly to both the Assistant District Director and the State Animal Health Official.

Testing

Diagnosing TB in live animals depends on using an effective testing technique with an intradermal injection of tuberculin obtained through your State animal health official or APHIS –VS Area Offices. Several varieties of tuberculin are produced. However, use only bovine purified protein derivative tuberculin (PPD bovis) licensed

by USDA for official testing. See table 1 for tuberculin test requirements for different species of animals.

Table 1 – Tuberculin test information for various animal species

Species	Dose and Type	Site	Read test visually and palpate
Cattle & Bison	0.1 mL PPD bovis	Caudal Fold	72 h +/- 6h
Horses	Not reliable		
Sheep & Goats	0.1mL PPD bovis	Caudal Fold	72 h
Swine	0.1mL PPD bovis	Base of Ear or Vulvar lips	48 h
Poultry	0.05 mL avian	Wattle	48 h
Exotic Bovidae	0.1mL PPD bovis	Midcervical	72 h
Deer, Elk & other Cervidae	0.1mL PPD bovis	Midcervical	72 h
Camelidae	0.1mL PPD bovis	Postaxillary Region	72 h

Note: *TB testing and test result interpretation for many exotic species (such as some zoo animals) are not yet developed or reliable. For interstate movement of these animals, contact the State animal health official in the State of destination for the TB-testing requirements (if any) for these species.*

Because the tuberculin test is based on an immune response, the animal being tested should not concurrently be receiving other medications, vaccinations, or anthelminthic drugs. These agents may temporarily affect the immune system and influence the result of the tuberculin test. This also means that sick animals may not be injected even if they are not being medicated or treated in any fashion. In addition, tail-bleeding is not recommended for other diagnostic procedures (e.g., brucellosis, Johne’s disease) at the time of tuberculin-test injection in cattle or bison because tail-bleeding may interfere with test interpretation.

Veterinarians are legally responsible for properly conducting and evaluating the results of tuberculin tests. Therefore, perform the test yourself; do not delegate the responsibility to a technician. For TB testing in species other than cattle or bison (e.g., cervidae), contact your State animal health official or APHIS –VS Area Offices for additional guidance.

Instructions for Testing

Step 1: Forms

1. Complete VS Form 6–22, Tuberculosis Test Record. (See Appendix D for an example of VS Form 6-22 Tuberculosis Test Record and instructions for completing it)
2. Identify the animal on the form by its official identification as outlined in the section entitled “Current Animal Identification.”

- All cattle and bison tested shall be individually identified by official eartags. Such identification must be recorded in its entirety on the test record at the time of injection and must be confirmed at the time of observation.
- Additional identification (such as bangle tags, non-official metal ear tags, neck chain numbers, tags, brands, horn numbers, and names) should also be recorded on the test record as supplemental information, but must never be used as the sole method of identification.
- When cattle and bison have been tagged with more than one official ear tag, all ear tag numbers must be recorded in their entirety.
- The breed, sex, and approximate age in years of each animal tested must be recorded in their entirety on the test record. Abbreviations such as C=Calf or A=Adult are not to be used.
- The owner should be informed of the number of animals injected, and advised to restrict them to the premises until the test is completed.

Step 2: Supplies

1. Tuberculin. Use USDA-Veterinary Services approved PPD Bovis tuberculin (see table 1). Check the expiration date to be certain that the tuberculin is still valid.
2. Syringe. Use disposable 1.0 cc plastic tuberculin syringe.

3. Needle. Use a 26-gauge, 3/8-inch-long needle; a larger gauge and longer or shorter needle might allow the tuberculin to leak from the injection site. A new needle must be used for each animal.

Step 3: Injection of Tuberculin

All cattle and bison tested must be sufficiently restrained to permit careful application of the tuberculin injection(s), correct reading of animal identification, and careful observation and palpation of the injection sites. No test should be applied or observed without having the animal restrained in a satisfactory manner. Nose tongs are no longer used by APHIS regulatory personnel.

1. In cattle and bison, injections should be made about 2 to 3 inches distal to the base of the tail. Rest the caudal fold on the forefinger exposing the area outside the hairline in bare skin near the center of the caudal fold.
2. Note scars, defects, and anomalies of the skin in this area on VS Form 6–22 so that they will not be confused with possible test reactions at the time of reading.
3. Use the caudal fold on either side of the tail; however, note which side you injected.
4. Clean the area to be injected, but do not use alcohol because it may be irritating to the skin.
5. Grasp the caudal fold between the thumb and index and middle fingers to stabilize it.
6. Carefully insert the needle to its full length between the superficial layers of the skin; withdraw it slightly and deposit 0.1 mL of tuberculin.
7. A small bleb should appear in the skin at the end of the needle.

Note: *It is important to establish a consistent injection technique (i.e., all animals should be injected on the same side of the tail)—particularly when testing large numbers of animals, unless there is some physical abnormality at the injection site.*

Step 4: Reading the Test

1. The test must be read between 66 and 78 hours after injection (72 hours is optimum). The veterinarian who made the injection must be the veterinarian who reads the test result. Exceptions must be approved in writing by the USDA, Veterinary Services, Assistant District Director. The veterinarian must

determine the results of the test by both observation and palpation of the injection site.
2. Verify the identification of the restrained animal and raise the tail to exert slight tension on the caudal fold.
3. Visually inspect the injection site closely and palpate it carefully to detect changes from the normal. Any swelling, sensitivity, or increase in thickness of the skin is considered to be a positive response to the tuberculin. The size of responses may vary and are not indicative of infectious status. Responses may be small, hard, pea-sized responses, diffuse responses, circumscribed responses, or large responses. If there is doubt about whether a response has occurred, palpate the opposite side of the tail to determine if there is a change from normal. Any observed change should be recorded.
4. Test observation without palpation is unacceptable.

Step 5: Recording the Results of the Test

1. Use VS Form 6–22.
2. Enter “N” (negative) when you observe no change in the tissue at the site of injection.
3. Enter “S” (suspect) when you observe or palpate any increase in caudal fold thickness, size, or sensitivity, at the injection site as described above.

Reactions and Interpreting Test Results

If an animal is exposed to the antigens present in bovine TB, a tuberculin injection results in a delayed hypersensitivity reaction manifested by swelling and induration at the injection site. A positive response usually begins within 8 to 12 hours and peaks about 72 hours after injection.

If the test produces any type of response, immediately notify your APHIS –VS Area Offices and State animal health officials. The caudal-fold test is used as a presumptive diagnostic procedure, and animals classified as suspect must be evaluated further by the comparative cervical (CC) test, gamma interferon test, or sent directly to slaughter under permit.

Only Federal or State regulatory veterinarians who have had specialized training may conduct the follow up testing. Follow-up CCT testing must be performed within 10 days of the initial caudal-fold injection in cattle and bison, or the herd owner must

wait 60 days (90 days for cervidae) after the injection of the CFT before a follow-up CCT can be administered. If the gamma interferon test is used as a follow up test the blood must be drawn within 30 days of the CFT inject date. Note: The gamma interferon test is not approved for use in Bison or captive cervidae. If the CCT or gamma interferon test indicates that the animal is a reactor, all further herd testing is conducted by Federal or State regulatory veterinarians.

Accredited veterinarians and regulatory veterinarians are expected to meet the caudal fold response rates as identified in the 2005 Bovine TB Eradication Uniform Methods and Rules, Appendix C. Improper injection or observation techniques, mishandling of tuberculin, and other events may lead to inaccuracies in the test and true disease status may be missed. See the Bovine TB Eradication Uniform Methods and Rules for more information. The most current version of this VS-published document is available on the APHIS website.

9 CFR part 77.1 Material incorporated by reference.

Uniform Methods and Rules—Bovine Tuberculosis Eradication. The Uniform Methods and Rules—Bovine Tuberculosis Eradication (January 22, 1999, edition) has been approved for incorporation by reference into the Code of Federal Regulations by the Director of the Federal Register in accordance with 5 U.S.C. 552(a) and 1 CFR part 51.

(a) The procedures specified in the Uniform Methods and Rules—Bovine Tuberculosis Eradication (January 22, 1999, edition) must be followed for the interstate movement of certain animals regulated under this part.

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