



MAY 20 2004

United States  
Department of  
Agriculture

**VETERINARY SERVICES MEMORANDUM NO. 552.16**

Marketing and  
Regulatory  
Programs

**SUBJECT:** Instructions and Procedures for Conducting Tuberculin Tests on Poultry and Swine

Animal and Plant  
Health Inspection  
Service

**TO:** VS Management Team  
Area Veterinarians in Charge, VS  
State Veterinarians

Washington, DC  
20250

**I. PURPOSE**

The purpose of this memorandum is to provide an outline of policies and standards for field stations and field personnel engaged in conducting tuberculin tests in poultry and swine.

**II. CANCELLATION**

This memorandum replaces ANH Division Memorandum No. 552.16, dated June 27, 1961, which is hereby canceled.

**III. GENERAL**

Animal species other than cattle, bison, and Cervidae may serve as reservoirs of bovine tuberculosis. Such reservoirs can thwart schemes to eradicate *Mycobacterium bovis*. The epidemiologic investigation of an affected herd should include careful study of these species of animals, with particular attention paid to those that were exposed to *M. bovis*. Appropriate actions include slaughtering, testing, and/or strict isolation of the exposed animals. This memorandum is a guide for testing poultry and swine for tuberculosis and for managing flocks and herds that are positive for the disease. Additional information for testing other susceptible species of animals is outlined in Veterinary Services Memorandum 552.15, dated April 2004.

**IV. INSTRUCTIONS AND PROCEDURES FOR CONDUCTING TUBERCULIN TESTS ON POULTRY**

Although not susceptible to *M. bovis*, poultry infected with *M. avium* may sensitize or infect cattle or bison.

**A. Administration of tuberculin test (avian)**

1. The following equipment is needed for administering the avian tuberculin test:
    - a. Small barrel tuberculin syringes with 26 gauge needles having 3/8 inch exposure of needle.
    - b. Wire, bottomless catching cage. Such equipment must be thoroughly cleaned and disinfected after use on each farm.
    - c. Brush, pail, and tuberculocidal disinfectant to use in disinfecting the catching coops, boots, etc.
    - d. Leg bands or marking brush for identification of reactor fowl. An alternate method of identification is to clip the tail feathers of reactors.
  2. Inject the avian tuberculin as follows:
    - a. Inject .05 cc of tuberculin into the wattle on the same side of each bird in the flock as birds are removed from the catching cage.
    - b. When catching cages are set up, it is easy to inject 100 percent of the birds, which is the most desirable level of testing in all flocks. Even if flocks have been regularly culled, reactors may be missed if a lower percentage of injection is employed.
    - c. Disinfect catching pens and boots between each premises.
    - d. Swelling of the injected wattle in 48 hours indicates a reactor. It is important that catching cages be used when making the observation to avoid missing reactors.
- B. Observation of clinical signs is sometimes used to detect avian tuberculosis instead of doing testing. (However, note that some birds may have a bone marrow form of avian tuberculosis and will react to the test without showing lesions. Therefore, use of the avian tuberculin test is recommended.) When observing flocks for clinical signs, follow these procedures:**
1. Use care and patience when entering a poultry house so as not to unnecessarily excite the birds. This will avoid, for example, death to layers from internally broken eggs (shell broken). A knock on the door of the laying house will alert the birds and your entry will not start their flying wildly in surprise.
  2. Where catching pens are not set up, a wire catching loop is a handy way to catch birds with clinical signs for examination.

3. Look for the following clinical signs of tuberculosis in poultry:
  - a. First observe the age of the chickens by noting feet, legs, presence or absence of spurs, pullet or old hen conformation, and feathering.
  - b. Lowered egg production and feed consumption should be noted.
  - c. If old hens are present, observe whether any are inactive, have pale combs and wattles, and bleached beak and legs.
  - d. Lameness and diarrhea are common in severely infected birds.
  - e. Perhaps the most pronounced clinical sign is the lack of flesh on the breast, sometimes described as "going light." Such birds are often caught very easily.
  
4. Demonstrate the gross lesions of tuberculosis to the owner using one or more birds showing such clinical signs. Do this carefully and neatly, using the following procedure:
  - a. Kill the chicken by "bleeding in the neck." This is done by holding the legs and wing tips in the left hand and grasping the head in the right hand. Rest the bird across your body; pull while twisting the skull back until the vertebrae are separated but the skin of the neck is not broken. This stretching thrust will permit the bird to bleed to death with the blood being held in the skin of the neck and not scattering about.
  - b. Next, place the carcass breast up on a clean surface; wet the feathers with the tuberculocidal disinfectant solution taken from the pail provided for washing instruments and hands. A knife and small rib shears are the instruments needed.
  - c. Remove a few feathers posterior to the breast, and place the feathers in a container.
  - d. Open the body cavity by incision, and cut the ribs with rib shears or small tin snips.
  - e. The breast may then be pushed upward to reveal the internal organs. Liver and spleen lesions are usually the most prominent; however, intestinal or generalized lesions are often apparent.
  
5. After the demonstration of lesions, the owner should burn the carcasses so they will not be scattered about or eaten by swine or other animals.
  
6. When lesions of tuberculosis are demonstrated, it is generally not necessary to administer the avian tuberculin test to the whole flock.

### C. Procedures for handling tuberculosis-affected flocks

In all cases, the elimination of avian tuberculosis is accomplished by sound well-established management principles. When avian tuberculosis is diagnosed in a poultry flock, the owner must be advised on the disposition of the fowl and on cleaning and disinfection as follows:

1. Infected flocks should be entirely disposed of by slaughter.
2. The owner should be asked to set an approximate date and to designate a preferred slaughtering establishment for disposing of the flock.
3. Buyers for slaughter should be asked to retain the identity of the farm of origin and report it to the inspector upon delivery of the birds to the slaughtering plants.
4. Small chicks may be kept on premises if sanitary arrangements are satisfactory. The owner should be advised that pullets and younger chickens are easily infected from contaminated ground, housing, and feed and water devices, as well as through mechanical transmission by attendants that take care of both the infected birds and the young stock. To help ensure that the eradication effort will be successful, it is recommended that all fowl on the premises be depopulated and that cleaning and disinfection procedures be diligently applied.

### D. Before chicks or pullets are again brought on the premises, the following actions should be completed:

1. Clean and scrape all droppings and litter from the poultry houses, roosts, and other equipment.
2. All feeding and watering equipment and nests should be removed from the house, emptied and thoroughly cleaned with a brush and soap and water, and then disinfected with an approved tuberculocidal disinfectant.
3. The equipment and as much as possible of the housing interior should be exposed to sunlight.
4. Any material (such as burlap, deteriorated insulating material, curtains, or equipment) that impedes cleaning should be removed and destroyed by burning.
5. The owner should be cautioned not to use litter and droppings to fertilize fields where livestock will forage or where drainage may expose other animals to infection. The combination of plowing under and exposure to sun will assist in the destruction of tuberculosis organisms present in the litter.

6. Apply lye solution. After the litter and debris are removed from the laying houses or shelters, the floors, roosts, and other equipment must be saturated with hot lye solution containing 1 pound of lye to 15 gallons of water. Let the solution remain for 24 hours if possible. After the lye solution has softened the dried manure, scrape again and flush with clean water. Note: **lye solutions are caustic.** Protect hands and face. None of the lye solution should be left available to children, livestock, pets, or poultry.
7. The entire house and equipment should then be sprayed with an approved cresylic acid disinfectant mixed at the rate of 4 ounces of disinfectant to 1 gallon of water. (Approved tuberculocidal disinfectants containing cresylic acid alone or in combination with phenolic compounds are available commercially.)
8. To avoid the undesirable odor of cresylic acid solutions, a solution of sodium orthophenylphenate may be used instead. Add 1 pound of sodium orthophenylphenate to 12 gallons of water at  $\geq 60^{\circ}\text{F}$ . If the temperature of the building is  $< 60^{\circ}\text{F}$ , the solution should be at  $120^{\circ}\text{F}$ , and even higher in very cold weather.

E. Poultry owners should be given the following advice:

1. Poultry should be confined to avoid cohabitation or contact with other livestock, such as cattle or swine.
2. Consider the many advantages of adopting an all-in, all-out production cycle with the purchase of pullets.
3. Do not keep laying flocks past their second laying cycle.
4. Raise all young birds for repopulation on clean ground, far enough away from old lots or farm yards so that young birds cannot get to the contaminated yards and old birds cannot contaminate the clean ground. In the summer, rearing may take place in movable summer shelters.
5. If permanent brooder houses are present, take care to prevent bringing in infection from old birds or yards through the caretaker's shoes. Place a pair of rubbers or overshoes in the brooder house to be put on upon entering and removed upon leaving.
6. Pullets on clean range should be culled and housed when they begin to lay eggs. They should be placed in previously cleaned and disinfected laying houses. They should never be in the same house as old hens.
7. When birds are housed, they should remain housed until they have completed their production cycle and are marketed. If hens, due to their profitability, are kept over beyond when the pullets are ready for housing, they must be housed separately.

## V. INSTRUCTIONS AND PROCEDURES FOR CONDUCTING TUBERCULIN TESTS ON SWINE

Swine are susceptible to *M. bovis*. They may become infected by association with infected cattle, bison, or other susceptible species and may transmit the disease to cattle and bison. Swine are also susceptible to *M. avium* complex mycobacteria and can, by shedding such organisms, expose and sensitize cattle, bison, and other susceptible animals so that they falsely respond to presumptive diagnostic tests for tuberculosis.

### A. In these two general situations swine might have to be tested for tuberculosis:

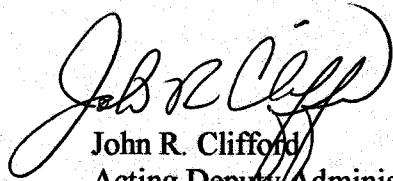
1. A designated tuberculosis epidemiologist has classified swine as exposed to tuberculosis by reason of association with an affected herd of cattle, bison, or captive Cervidae. In such a case, it is preferable to depopulate rather than test the exposed swine. There are provisions for indemnity if the swine are depopulated; however, a herd plan may include a requirement that the swine be tested for *M. bovis*.
2. A swine herd not associated with a known affected herd of cattle, bison, or captive cervids is suspected of harboring *M. bovis* or *M. avium* complex due to clinical signs or the detection of tuberculous lesions in swine slaughtered or necropsied. In such cases, testing may provide useful information for understanding the epidemiology of the disease in the herd and the possible source of exposure.

### B. Procedures for conducting tuberculin tests on swine

1. Restraint
  - a. The speed and ease of testing will depend largely on available handling facilities. Many prefer a wire-loop catcher or the use of swine-catching crates. However, if the animals to be tested can be handled in an area where they are regularly kept (such as a feeding floor), they will not be as restless and nervous as when put into a strange pen or area. Most sows are easily injected by crowding one or more into a corner with a solid panel. In the interest of safety, special precautions should be used to handle boars. When large numbers of animals are to be tested, it is preferable to use long narrow chutes with drop gates.
  - b. Advise the owner to keep all swine to be tested penned away from mud holes starting the day before testing is to begin. In most instances only breeding stock or animals intended for breeding need to be tested.

2. Injection technique
  - a. Since swine are susceptible to both avian tuberculosis (*M. avium*) and mammalian tuberculosis (*M. bovis*), suspected swine are injected with both types of tuberculin.
  - b. Sites of injection are the lips of the vulva and the soft skin just posterior to the ear. Boars may be injected at the junction of the skin and mucus membrane of the anus. Use *M. avium* on one side and *M. bovis* on the other. Clean the site of injection with dry cotton or fiber toweling when necessary.
  - c. Syringes should be disposable 1.0 cc and/or 0.5 cc tuberculin syringes. The needle should be 26 gauge with 3/8 inch exposure. The dosage is 0.1 cc. injected intradermally. Identify the syringes as to type of tuberculin it contains by wrapping a piece of tape around the barrel. The needle, chuck, and syringe are maintained in a clean condition by swabbing with a pledget of cotton saturated in alcohol.
3. Identification of swine. Unless the swine are previously identified by ear tags or special marking, they should be ear tagged at the time of injection.
4. Observation and interpretation of the test
  - a. The swine should be observed 48 hours after injection.
  - b. Swine with tissue reactions that can be observed or palpated should be classed as reactors, being careful to note whether the animal is reacting to *M. avium* tuberculin, *M. bovis* tuberculin, or both.
  - c. Care must be used in observing females that may be in estrus when the injection site is the vulva.
5. Management of infected swine and affected swine herds
  - a. Swine classified as exposed to *M. bovis* by reason of association with a tuberculosis-affected cattle, bison, or cervid herd may be eligible to be depopulated with indemnity under 9 CFR 50.3.
  - b. Negative animals from herds containing valuable breeding lines may be immediately segregated to clean quarters and retested at 60-day intervals until two negative tests are obtained following slaughter of the infected group. However, because false negative test results are possible, this test-and-removal approach should be discouraged.
  - c. Feed sacks, shovels, scrapers, or other tools or material used for poultry must not be used for swine.
  - d. The extermination of rats or other rodents is an essential part of the tuberculosis eradication program.
  - e. If feral swine are in the area, their tuberculosis status should be determined, if possible, and they should be prevented from contacting domestic swine herds.

- f. A complete epidemiological study of each affected herd must be conducted and reported.
- 6. Cleaning and disinfection and segregation
  - a. All infected structures such as hog houses; feeding and breeding floors should be thoroughly cleaned and disinfected with an approved tuberculocidal disinfectant.
  - b. Infected soil areas should be fenced off and kept free of animals and fowl for at least one cropping season.
  - c. Poultry must not be permitted to mingle with swine under any circumstance; swine should not be put on ground, or in other areas, formerly populated by poultry.
  - d. Swine must not be fed dead poultry, uncooked garbage, offal, or contaminated milk or milk products. Swine feed must be free of contamination by poultry or other swine.



John R. Clifford  
Acting Deputy Administrator  
Veterinary Services