

DIRECTIONS FOR USE

PULLORUM ANTIGEN STAINED ANTIGEN POLYVALENT

FOR VETERINARY USE ONLY – CONTAINS FORMALDEHYDE AS A PRESERVATIVE

Recommendations for use: This product contains standard, intermediate, and variant strains of *Salmonella Pullorum*. The bacteria are inactivated and therefore, cannot spread disease. The product is designed for use in the whole blood or serum plate agglutination (SPA) tests for the detection of pullorum disease and fowl typhoid. When the antigen is mixed with blood or serum samples of infected birds, it reacts to produce clumping with the blood or serum.

Indications for use: The antigen is used in a screening test for the Pullorum status of a flock. The antigen should not be used as a final determination of the infection status of individual birds. When used on a flock basis, it provides a simple method for detecting the presence of antibodies in the population. As in all serologic assays, positive reactions should be followed by other tests to confirm the infection status of the flock. It is strongly advised that flocks with positive reactions be sampled for confirmatory testing before the infection status is verified. Repeated testing is also needed to continue certification that the flock is negative. Consult your poultry veterinarian in case of any clinical signs or questionable reactions. **DO NOT DESTROY ANY FLOCK BASED UPON RESULTS OF TESTING WITH THIS ANTIGEN.**

Performance of the test: Since it is a simple, rapid screening test, the SPA and whole blood assays have limits for sensitivity and specificity. Confirmatory testing should be used if positive reactions occur. For pullorum disease, a culture of the organism is often used to confirm positive samples. Culturing of the organism is available from Charles River or any NPIP approved laboratory. Other diagnostic tools such as PCR should also be used to confirm serologic reactions. Rely on your veterinarian and diagnostic laboratory professional for guidance.

Directions for use: ALWAYS USE THE ANTIGEN UNDILUTED, AND ALLOW TO COME TO ROOM TEMPERATURE BEFORE USE.

Conducting the whole blood test:

1. Shake the antigen well.
2. Place a drop of antigen on a test plate with the dropper contained in the kit.
3. Draw blood from the test bird by lancing the vein under the wing.
4. Using the loop supplied with the antigen, administer a loop-full of blood to the antigen drop on the plate.
5. Mix the blood with the antigen by stirring with the loop. Pick up the plate and rotate it several times for further mixing.
6. The reaction is read at the end of the 2-minute incubation period. A positive reaction shows formation of discrete clumps of stained material, normally starting at the edge of the mixture. Negative reactions show little to no change in the opaque serum antigen mixture.
7. Rinse the loop with clean water and dry between tests to prevent cross contamination.

Conducting the SPA test:

Required Components: Pullorum Plate Antigen; positive and negative control sera; test samples (never frozen); a glass plate or mirror marked in squares of ~1-1.5in (~25-40mm); pipettes; clean water

Procedure:

1. Allow all components (solid plate, antigen, control sera, and test sera) to warm to room temperature before use.
2. Shake the antigen well to mix. It is recommended to use a vortex-type mixer for 15 to 20 seconds.
3. Place one drop of antigen in each square on the glass plate or mirror, using the dropper contained in the kit. One drop is administered per sample or control.
4. It is advisable to test known positive and negative control sera at the start of all testing. Use separate pipettes for administering all controls and samples. Pipette one drop (approximately 0.04 to 0.05ml) of positive control serum in one square, and one drop of negative control sera in another. Place one drop of each test serum in individual squares.
5. Mix the serum with the antigen by pipetting up and down when administering each control or sample. Be sure to keep each mixture within their own square.
6. Incubate at room temperature for 2 minutes.
7. The plates may be rotated by hand during incubation. Gently rotate the plate for a few seconds. Let stand. After 1 minute, the plate may be rotated again and allowed to stand.
8. The reaction is read at the end of the 2-minute incubation period. A positive reaction shows formation of discrete clumps of stained material, normally starting at the edge of the mixture. Negative reactions show little to no change in the opaque serum antigen mixture.

References:

1. Beard, C.W., 1989. Chapter 44, Serologic Procedures. In: A Laboratory Manual for the Isolation and Identification of Avian Pathogens. 3rd edition. American Association of Avian Pathologists. University of Pennsylvania, Kennet Square, PA.
2. National Poultry Improvement Plan and Auxiliary Provisions Manual, USDA APHIS.

Precautions: Avoid exposing hands, eyes or clothing to the antigen. It is recommended that the test be conducted out of direct sunlight, where there is a minimum amount of dust. For product or technical information, please call Charles River Customer Service at 1-860-889-1389.

Packaging: The antigen is packaged in a glass vial with a separate dropper tip and loop. The dropper is calibrated to deliver 0.04ml of antigen per drop (one test). The antigen should be stored in the dark at 2 to 7°C (35-45°F). **DO NOT FREEZE.**

How Supplied:

40 ml – 1000 tests	Catalog No. 10105346
14 ml – 350 tests	Catalog No. 10102217

Supplemental Reagents:

Pullorum control serum	Catalog No. 10100702
Negative control serum	Catalog No. 10100511

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