

Pullorum and Paratyphoid in North Dakota Turkeys

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Poult mortality is the cause of heavy losses to turkey producers in North Dakota. The formation of the Turkey Improvement Plan based somewhat on the National Poultry Improvement Plan has done much to increase the interest of turkey producers and hatcherymen in the control of pullorum disease in these birds. During 1944 and 1945 the North Dakota Agricultural Experiment Station, in co-operation with the North Dakota Poultry Improvement Board,⁴ and the Agricultural Extension Service has conducted investigations in an effort to determine the incidence of pullorum disease in the turkey flocks of the state. The studies have included various methods of detecting agglutinins for *S. pullorum* in whole blood and serum and the isolation and identification of organisms obtained from poults and mature turkeys.

During the winter and early spring of 1944-45 field tests of prospective breeding flocks were made. The usual procedure was for the testing and selecting crews to test the blood by means of the whole blood plate test in which the K antigen⁵ was used. At the same time that the field test was made, additional blood was drawn and sent to the laboratory where serum plate and tube tests were run. In the field tests one standard loopful of blood and one drop of antigen were used and the readings made at two minutes. In the laboratory the serum plate test was made with 0.02 cc. of serum and 0.05 cc. of antigen. The readings were made at two minutes and again later with notations made as to the time of reaction.

The plate tests were made using K antigen. The tube tests were made with a finding dilution of

1:20 using either commercial T. G. antigens or antigens made from the same stock cultures as were used in the K antigen. The antigens were prepared according to directions given in the bulletin (1) describing the National Poultry Improvement Plan. All readings of tube tests were made after 18 to 24 hours incubation at 37° C. In nearly all cases reacting sera were again tested using 1:20, 1:50 and 1:100 dilutions. Few comparative data are presented on the tube and serum plate tests because of inability to obtain sufficient numbers of satisfactory samples. Under the conditions encountered it was frequently very cold when samples were drawn and in many cases there was more or less hemolysis. This does not appear to interfere with the serum plate test, but it makes the tube test very difficult to interpret, especially in low dilutions.

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⁴The authors wish to express their appreciation to the North Dakota Poultry Improvement Board for funds used in paying some of the expenses of this study.

⁵The authors wish to thank Dr. H. W. Schoening of the U.S.D.A. Bureau of Animal Industry for furnishing a part of the K antigen and the directions for preparing the antigen used in some of the comparative tests.

The authors are indebted to Nora Edlund for technical assistance in these studies.

In the diagnosis of pullorum in young turkeys the lesions, history and identification of organisms have all been considered but no diagnosis of pullorum disease has been made, unless small Gram negative rod-shaped organisms that formed acid and gas in dextrose but failed to change sucrose, maltose, or lactose were isolated. In cases of doubt concerning the identity of some cultures, samples were sent to the Salmonella Laboratory at the University of Kentucky.⁶

Results of Agglutination Tests

The results of the agglutination tests were quite similar to those reported by other workers. The whole blood plate gave by far the fewest reactions, the serum plate test and the tube test agreed fairly well provided the plate test was read at five minutes instead of the usual two minutes and provided the tube dilution did not exceed the 1:50 dilution.

This observation confirms the opinions expressed by various workers (2) (3) (4) as regards the value of the whole blood test as it is routinely used. Dickinson, Rosenwald and Morrill (5) have previously reported fair agreement between the serum plate and tube tests for detecting pullorum disease in turkeys.

Examination of Poults

It was found quite early in this study that the incidence of pullorum disease in this area was quite low. As shown in the report of the diagnostic laboratory (6) there were only nine cases of pullorum in poults found in the period of July 1, 1943 to June 31, 1944. Four of these cases originated outside this state. Similar results were found during the present brooding season. Out of 63 lots of poults composed of 255 birds there have been 6 cases of pullorum while paratyphoid cases numbered 11 and navel infection 30 cases. These data indicate quite clearly that pullorum

is not the most important disease in North Dakota turkeys. The losses from paratyphoid infection have in many instances been quite high but navel infections have been by far the most serious cause of poult mortality in 1945. Of the six cases of pullorum disease that were found, one originated in North Dakota, one in Minnesota and the other four in Iowa.

Discussion

An interpretation of the data collected in this investigation strongly indicates that an effort should be made to control paratyphoid infection and at the same time keep pullorum disease at a low incidence. Hinshaw and McNeill (7) have shown that the incidence of paratyphoid can be greatly decreased by the use of a proper type of antigen with the tube test. These same authors (8) have shown that non-specific agglutinins can seriously interfere with a standard pullorum test. Sanders, Pomeroy and Fenstermacher (9) had previously found that many Minnesota poults were infected with paratyphoid infections and that the whole blood agglutination test was unsatisfactory both for the detection of pullorum disease and the various paratyphoid infections.

As one means of decreasing poult losses it is suggested that the breeding flocks be tested at least twice using the serum plate test and the K antigen with the reading time of five minutes and that at the time of the second pullorum test the sera be tested with an antigen made from *S. typhimurium* and that the reactors to the paratyphoid antigen be removed from the flocks. The reactors to paratyphoid need not be considered in the classifications of the pullorum control plan.

In only a few instances have examination of reacting mature turkeys been made. This phase of the study was not stressed because of the few cases of pullorum disease

⁶The authors wish to thank Dr. Dimock and his staff for identifying these cultures.

encountered in poult and the lack of agreement among the various agglutination tests employed. A few examples of typical cases can be used to illustrate some of the problems to be considered in a pullorum disease control program.

Flock I—Tested with a commercial antigen April, 1944. History of heavy losses in mature birds and poults. Of 85 tested 38 were positive, 2 were suspicious and 45 negative. A reacting bird was examined at post-mortem and a culture of *Shigella gallinarum* isolated from the ovary. The flock was disposed of before other tests could be made. This flock was tested with a commercial antigen.

Flock II—This flock had been vaccinated several times using the cholera-typhoid bacterin. The flock was tested December 6, 1944 at which time 784 birds were tested. Five birds reacted to the whole blood plate test. Two hundred fifty-five sera samples gave suspicious to markedly positive reactions to the plate tests. It was impossible to make accurate readings of the tests because the reactions frequently occurred after as long as 15 minutes. The tests were made on plates covered with a raised glass cover that retarded evaporation and allowed for timing of the tests, without rapid drying. The serum plate tests and 1:20 dilution of the tube tests agreed very well. There did not seem to be any correlation between the rate of reaction on the serum plate test and the titre of the serum. Many samples that were marked suspicious at 15 minutes gave a complete agglutination at 1:50 dilution in the tube test.

From the data obtained on vaccinated flocks in this study it is suggested that no tests be made on birds that have been inoculated with a typhoid bacterin for a period of at least two months.

Flock III—Eleven birds, all of which had been vaccinated with a cholera-typhoid vaccine 2 months

previous to testing. One reacted to all three tests. The bird was killed and samples for culturing taken from the testes, liver, long bones and peritoneal cavity. A single staphylococcus culture was obtained.

Flock IV—A flock with a history of no pullorum or other serious infectious diseases. The poult mortality the previous spring had been extremely low. This flock was tested November 27, 1944. At this time 421 birds were tested with one reacting to the whole blood plate test and 89 reacting to one or both of the serum tests. On December 13, 1944 three reacting were tested again with all reacting to the plate tests and all at 1:20 and 2 at 1:50 dilution with the tube test. The birds were killed and samples taken for culturing. No organisms were obtained. On January 21, 1945, 25 reacting birds were retested. At this time 2 reacted to the whole blood test, 7 to the serum plate and 5 to the tube test.

It was concluded that the flock was not infected with pullorum disease and no evidence of pullorum disease has been found in poults from this flock.

Flock V—This flock had a history of poult losses due to pullorum disease in 1944. The first test was made December 2, 1944. The samples were all hemolyzed so no tube tests were made. There were 340 in the flock, but only six reacted to the whole blood test, while 25 gave a "suspicious" to strongly positive test by the serum plate method. The reactors were removed from the flock and a retest made January 3, 1945. At this time only one suspicious reactor to the whole blood test was found and this bird failed to react to either the serum plate or tube tests.

There were, however, nine positive and three suspicious samples with the serum plate test and 12 positive with the tube test. The data are as follows:

	W.B.	Serum Plate	Tube 1:20
1	—	+	+
2	S	—	—
3	—	+	+
4	—	+	+
5	—	S	?H
6	—	+	+
7	—	S	—
8	—	S	+
9	—	+	+
10	—	+	+
11	—	+	+
12	—	—	+
13	—	—	+
14	—	+	—
15	—	+	+

H—Hemolyzed

This flock is to be studied further. The indications are, however, that a single test will not remove all potential reactors to the plate or tube tests in flocks with a known pullorum infection

Summary

1. Various types of agglutination tests for the detection of pullorum disease in turkeys are discussed.
2. The serum plate test with the reading at 5 minutes is recommended.
3. Pullorum disease is not widespread in North Dakota turkeys.
4. Both paratyphoid infections and navel ill are responsible for greater losses than pullorum disease in North Dakota poults.
5. It is suggested that an effort be made to decrease the incidence of paratyphoid infection by means of a testing and elimination program.
6. Some of the factors responsible for reactions to the pullorum test are discussed.

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