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Service

Veterinary Services

**Epidemiologic and Other Analyses of  
Indiana HPAI/LPAI-Affected Poultry Flocks:  
March 18, 2016 Report**

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## I. EXECUTIVE SUMMARY - UPDATED

In mid-January, a combined outbreak of highly pathogenic avian influenza and low pathogenic avian influenza (HPAI and LPAI) occurred in Indiana. The outbreak in commercial turkeys was first detected by an increase in mortality followed by laboratory confirmation of H7N8 HPAI. After initial efforts to control the disease, a series of epidemiologic, geospatial, genetic, and wildlife investigations was started. These studies are being conducted collaboratively with the poultry industry, the Indiana State Board of Animal Health, and the United States Department of Agriculture's (USDA) Animal and Plant Health Inspection Service (APHIS). This is the second report of the findings available to date to assist in understanding the introduction and disease transmission pathways. Ultimately, the goal is to reduce the risk of continued spread in this outbreak and to help in future efforts to prevent disease incursions. This report is the final epidemiologic report for this incident, although additional analyses may be undertaken and reported through other mechanisms.

Genetic analyses determined that all H7N8 viruses detected from this event are of North American wild bird lineage, and the HPAI and LPAI viruses are highly similar across all eight genes excluding the multi-basic amino acid insertion at the cleavage site in the HPAI virus. Genetic analysis also suggests a point source introduction followed by lateral/secondary spread.

An investigation of cases and controls using an in-person administered questionnaire examined physical and management characteristics of infected premises. Differences were identified between case farms and barns compared to control farms and barns. Several risk factors found in a previous HPAI case-control study were found to occur on more case farms and in case barns than on control farms and in control barns. These factors included distance to dead bird disposal and litter compost areas, presence of wild mammals, and visitors entering barns.

Additionally, daily mortality sheets were obtained from both case and control farms. Mortality patterns from these farms between September 7, 2015, and January 17, 2016, were compared. This comparison showed differences in mortality between case and control farms for 2 weeks in October 2015 and again in November 2015. However, there were no major differences in mortality patterns between case and control farms in closer temporal proximity to the identified H7N8 HPAI outbreak.

APHIS has completed sampling and testing of wildlife near infected premises. The results indicate no evidence of existing virus in the samples from wild birds and mammals tested.

Initial geospatial analysis looked at county-level factors that may have contributed to the introduction of the virus into Dubois County, Indiana. The weather in Dubois County was warmer and wetter than past years, which may have contributed to the introduction and persistence of the virus. Additional analysis identified a geospatial pattern observed between infected premises and a driving route. More detailed geospatial analysis is ongoing.

## II. INTRODUCTION - UPDATED

In 2015, the United States experienced an outbreak of highly pathogenic avian influenza (HPAI) that was the largest animal health emergency in history. Many lessons were learned from that experience, including the need to understand the outbreak to help prevent and control future outbreaks. Since the end of the outbreak in the summer of 2015, the United States Department of Agriculture's (USDA) Animal and Plant Health Inspection Service (APHIS), States, and the poultry industry have been preparing on many fronts for potential outbreaks in the fall of 2015 or in 2016. The January 2016 combined outbreak of HPAI and low pathogenic avian influenza (LPAI) in Indiana has challenged the preparations all the groups have made. In response to the outbreak, APHIS Veterinary Services (VS) – in collaboration with the Indiana State Board of Animal Health and the southern Indiana poultry industry – has undertaken a number of epidemiologic, geospatial, and laboratory-based investigations.

These investigations include:

- A field-based study of cases and controls using data collected through personal interviews with farm personnel;
- geospatial analyses;
- on-farm sampling of wildlife;
- phylogenetic investigations; and
- an assessment of mortality data

This report updates the previous summary of preliminary findings from these studies. Although this report is intended to be the last epidemiologic report for this outbreak, additional analyses will be completed to provide information to producers, industry, and other stakeholders on how the disease may have been introduced and spread to help with current and future mitigation efforts.

### III. GEOSPATIAL ASSESSMENT FOR H7N8 HPAI/LPAI OUTBREAK - UPDATED

APHIS completed a county-level analysis on available data for Dubois County, Indiana, and surrounding counties. The focus was on descriptive epidemiology that could explain why Dubois County experienced an HPAI and LPAI outbreak of H7N8 in early 2016 based on available geospatial data. The analysis focused on the introduction of H7N8 LPAI by wild waterfowl and environmental conditions that may have or have not supported this introduction.

#### A. Highlights of this Analysis

- The mean high and mean low temperatures for the 6 weeks leading up to the outbreak were warmer than the previous 2 years and were conducive to Influenza A virus survival.
- Precipitation in 2016 in the 6 weeks leading up to the outbreak was higher than in 2015, but lower than 2014. However, when paired with generally warmer temperatures, the environment was still likely conducive to support survival of H7N8.
- Dubois County is in the same watershed as and downstream from a large reservoir and the Hoosier National Forest, possible sources of LPAI virus introduction into the area.
- Presence of cropland in Dubois County did not appear to be a factor that influenced the presence of H7N8 LPAI-infected waterfowl relative to other counties.
- Dubois County was at the lower range for sources of open water in relation to other counties. Therefore, open water is not considered a factor that may have increased the presence of infected waterfowl in Dubois County compared to other counties.
- Dubois County has one of the higher densities of poultry compared to surrounding counties and therefore had a larger susceptible population for introduction of LPAI into commercial poultry.
- There was a strong pattern observed between infected premises and a common driving route, which followed a general north-to-south direction.

#### B. Variables Considered for Geospatial Analysis

Multiple variables were assessed geospatially to help explain why the avian influenza virus may have been present in the Dubois County area in the late fall/early winter of 2015-2016. Table 1 below outlines the variables and their respective findings.

**Table 1. Variables evaluated geospatially**

| Measured Variable      | Observation   | Notes  |
|------------------------|---|--|
| Outdoor Temperature    | Mean high and low temps higher  | Evaluated 6 weeks prior to first detection; Compared to past 2 years             |
| Precipitation          | Total precipitation higher in six weeks prior to outbreak than previous year, yet lower than in 2014. | Evaluated 6 weeks prior to first detection; Compared to past 2 years             |
| Watershed connectivity | Watershed connectivity to Hoosier National Forest and nearby large reservoir                          | May have allowed for introduction of LPAI into the area via connected watersheds |

| Measured Variable                               | Observation  | Notes  |
|---|--|--|
| Presence of cropland                            | Presence or absence of cropland in the area was not a factor | Data for specific crop type(s) near/around the infected premises was not available and therefore not evaluated               |
| Open water source                               | Open water source was not a factor                           | ...  |
| Poultry density                                 | Higher poultry population provided a susceptible population  | Dubois County did have a denser poultry population than surrounding counties   |
| Elevation of operation                          | Had some association with disease                            | Limited number of infected premises could not lend statistical significance and was confounded with the location of the road |
| Roads – distance and direction                  | Had likely association with disease outbreak                 | Strong pattern observed between infected premises and common driving route   |
| Barn directional orientation                    | Orientation (N,S,E,W) was not a factor                       | No observable pattern among or between infected and uninfected barns in the control zone                                     |
| North-to-south orientation of infected premises | No discernable explanation at this time                      | A greater number of infected premises may have lent more ability to statistically evaluate variables related to this         |

**C. Temperature and Precipitation**

Temperature and moisture are factors that influence migration patterns in some waterfowl (Austin et al., 2002). Temperature and moisture for a 6-week period prior to detection in January 2016 were compared to the same period in the previous 2 years. The mean high temperature in 2016 was higher than 2 previous years (Figure 1). The mean high was 50.88 degrees Fahrenheit and the mean low was 34.04 degrees Fahrenheit—above freezing temperatures (Figure 2). These temperatures have been shown to be conducive to persistence of avian influenza virus in the environment. Precipitation in the 2016 period was higher than 2015, but lower than 2014 (Figure 3). However, when paired with temperature information, the moisture in 2016 may have created an environment conducive to H7N8 LPAI survival and the presence of waterfowl.

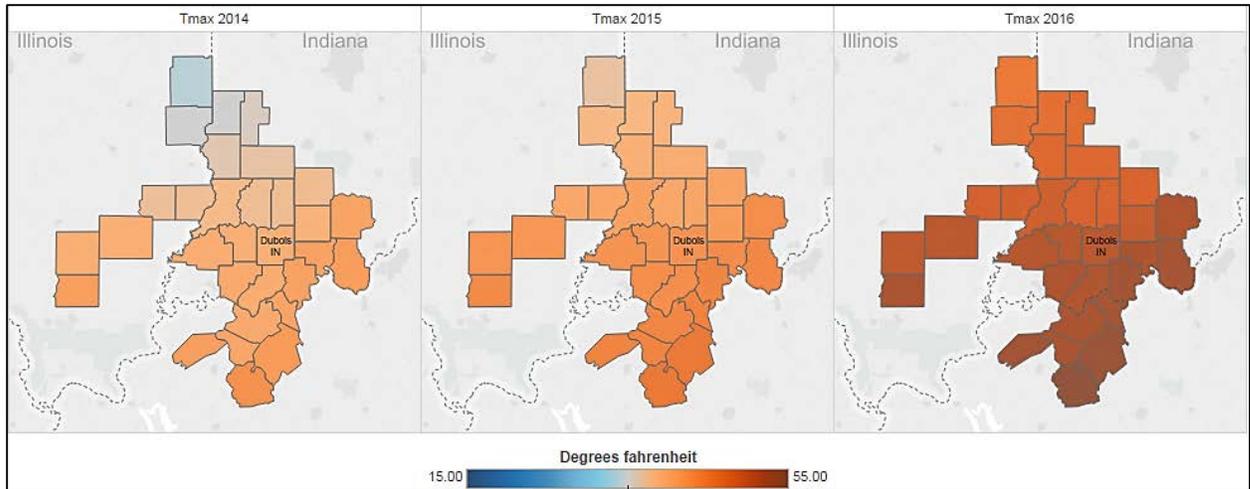


Figure 1. Average daily maximum temperature by year for a 6-week period of December and early January.

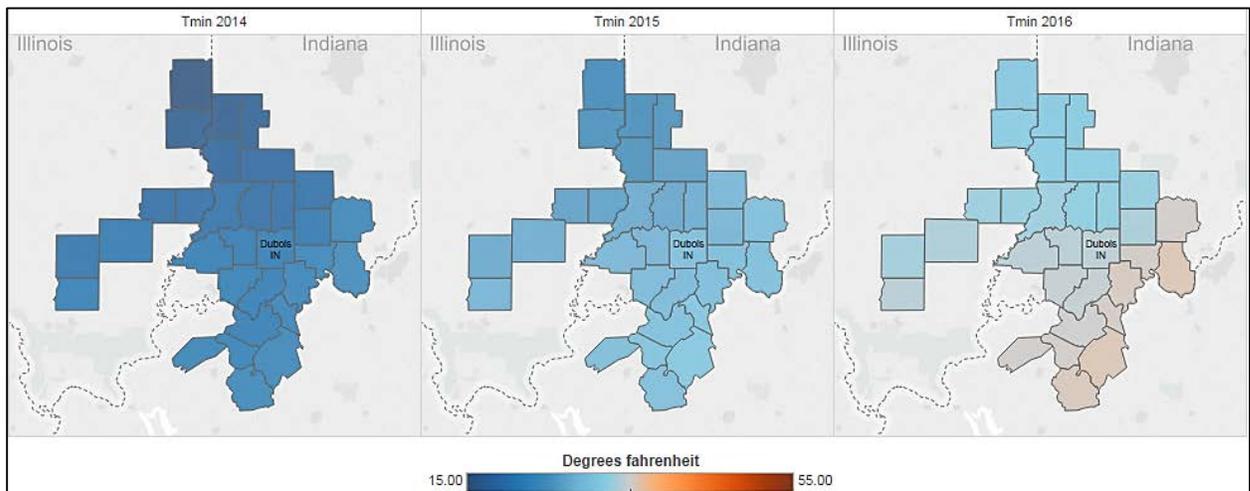


Figure 2. Average daily minimum temperature by year for a 6-week period of December and early January.

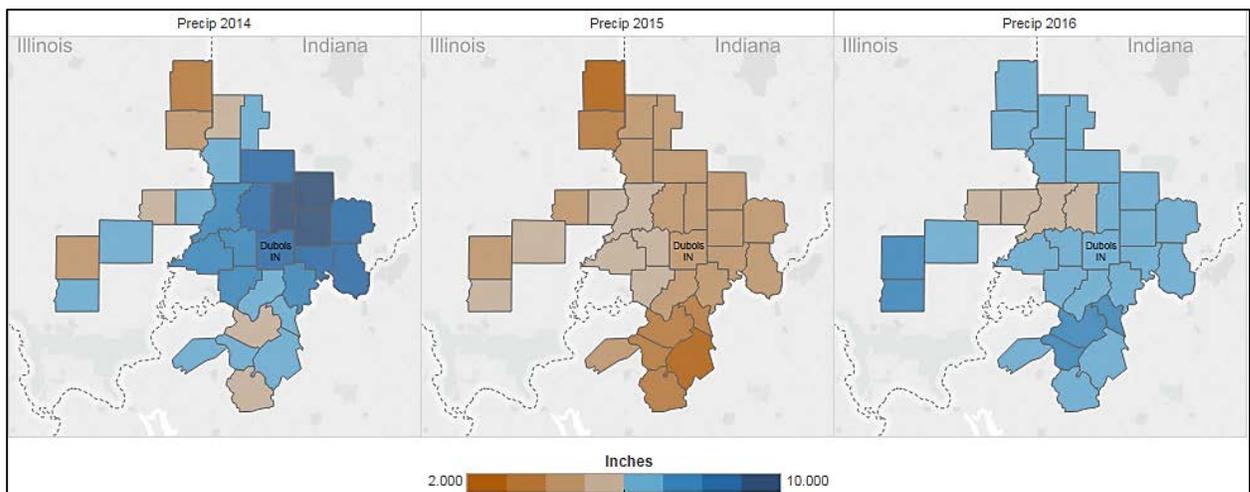
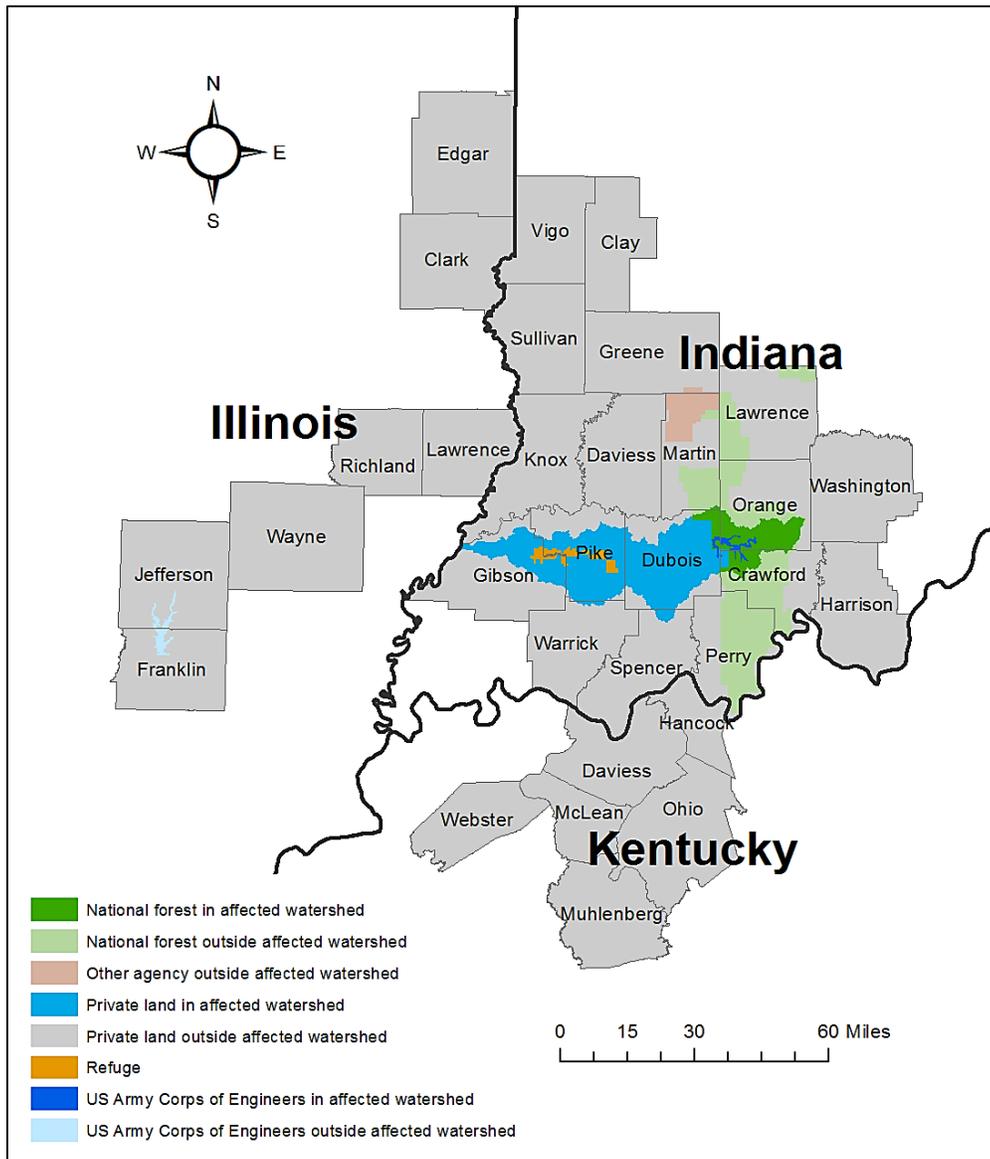


Figure 3. Total precipitation by year for a 6-week period of December and early January.

### D. Federal Land and Watershed

Avian influenza viruses have been isolated from water where waterfowl congregate, even after the birds have moved (Ito et al., 1995). Hoosier National Forest is the largest contiguous land area in Indiana that is unaltered by agriculture or urban development (Leatherberry, 2002). Habitats such as Hoosier National Forest create important areas for resting, feeding, or other uses for migratory waterfowl (Kaminski et al., 1989; Leatherberry, 2002). The watersheds that included all 10 infected premises also were hydrologically connected with Hoosier National Forest (Figure 4). Favorable conditions (see Temperature and Precipitation section) within a watershed may attract waterfowl during winter months (Kaminski et al., 1989). If waterfowl were to follow the watershed, or if heavy rains washed feces from infected waterfowl in Hoosier National Forest downstream to Dubois County, these conditions could create an opportunity for introduction into Dubois County that was not shared with other counties in Indiana.

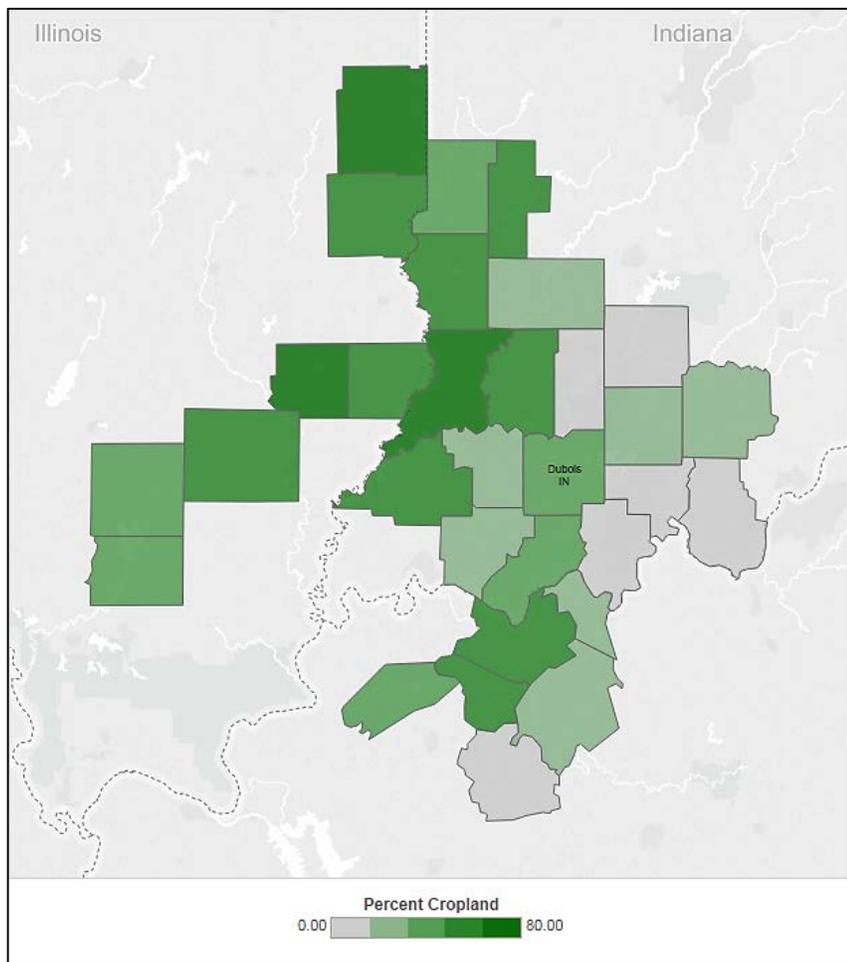


**Figure 4. Dubois County is downstream from and within the same watershed as Hoosier National Forest.**

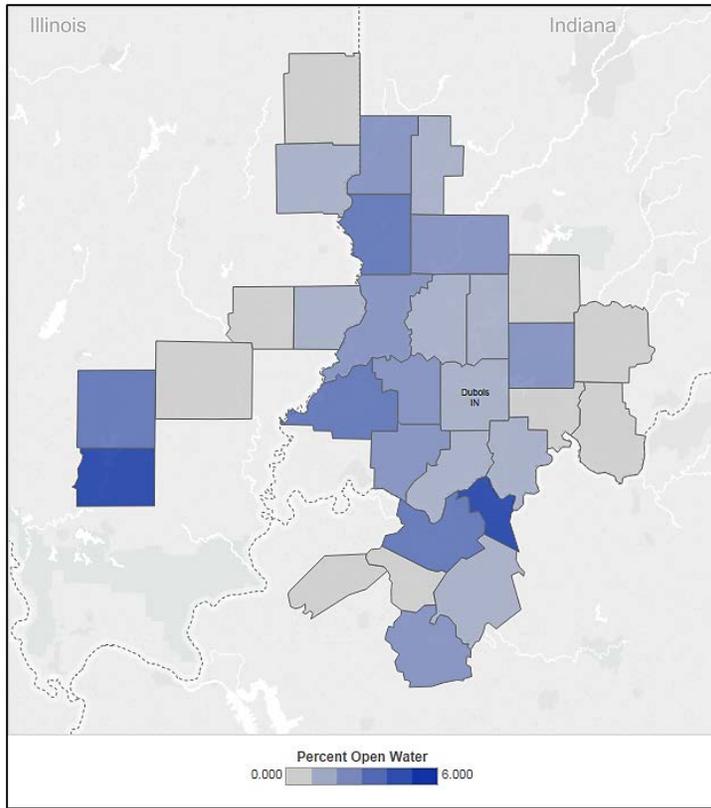
**E. Other Attributes**

An assessment of H5N2 HPAI in Minnesota indicated the presence of cropland may contribute to the presence of infected waterfowl. Cropland is an important component of migratory waterfowl habitat (Kaminski et al., 1989). In Indiana, the percentage of cropland in the counties studied ranged between 3.5 and 78 percent. Cropland did not appear to be a factor that influenced the presence of H7N8 LPAI-infected waterfowl in Dubois County relative to other counties (Figure 5). Additional analysis of water usage within crops, the type of crops, and proximity to infected premises may provide additional information.

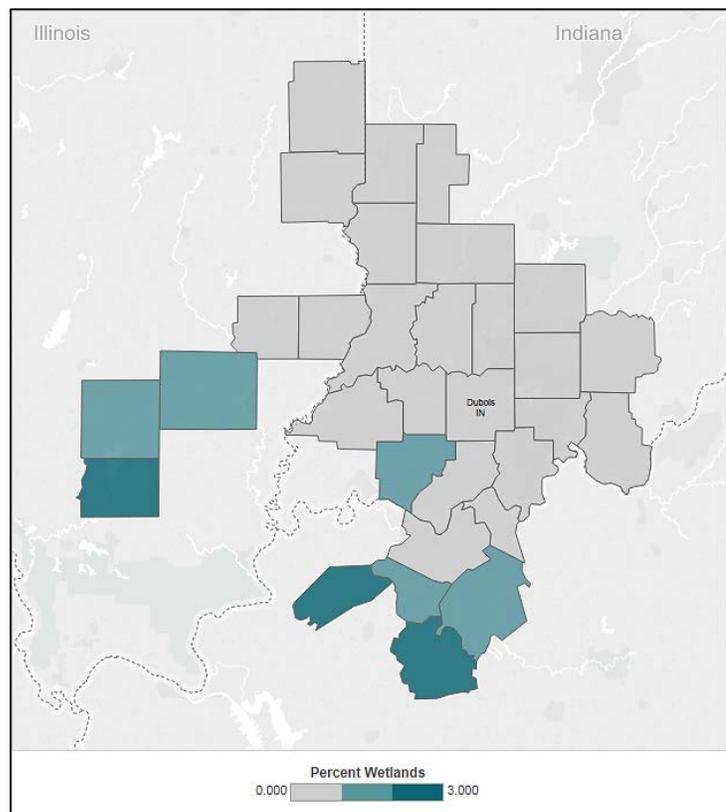
The percentage of wetland and open water was also explored as possibly important for waterfowl migration (Bellrose and Trudeau, 1987). Dubois County was again in the lower range of the spectrum and these attributes were not considered factors that influence the presence of infected waterfowl in Dubois County relative to neighboring counties (Figure 6 and Figure 7).



**Figure 5. Percentage of county comprised of cropland. 2014 NASS Cropscape.**



**Figure 6. Percentage of county comprised of open water (2014 NASS Cropscape).**



**Figure 7. Percentage of county that is comprised of wetland (2014 NASS Cropscape).**

### F. Roads and Elevation

Two geospatial assessments were conducted to address the apparent association of roads to reported cases. The first assessment, also known as a connectivity test, evaluated the direct connection between cases and a common network of north-to-south roadways, heretofore referred to as the connecting route. The connecting route consists of four different roads: Highway 545, East Jasper Dubois Road, North Steinbottom Road, and Highway 164 (Figure 8).

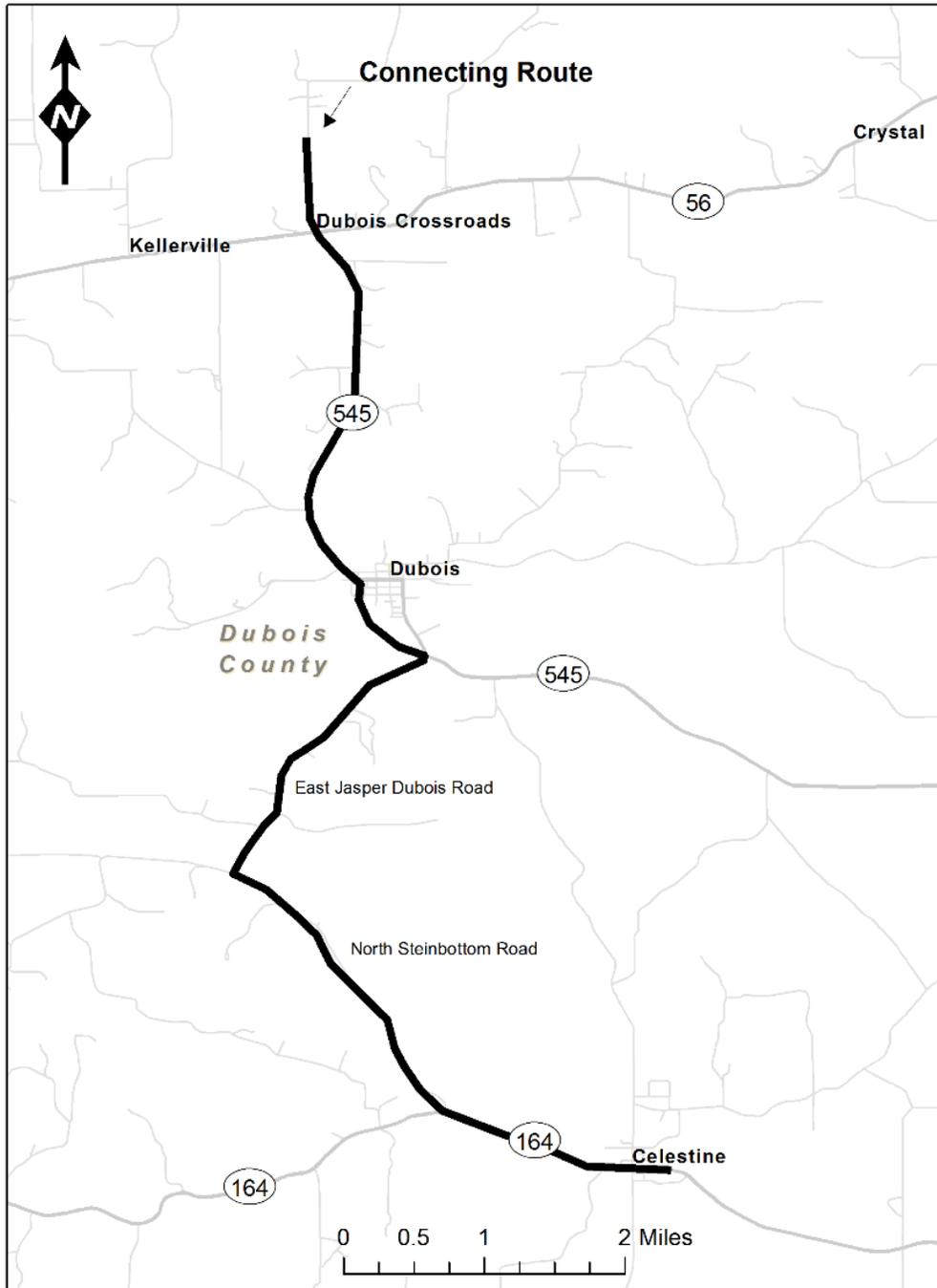


Figure 8. The connecting route consists of four different roads.

The second assessment compared distance of cases to the connecting route. A distance of 500 meters was selected to compare results with previous AI outbreak studies (personal communication, Malladi, S., Weaver, J.T.). Comparisons were made between those premises that were within 500 meters from the connecting route and those premises that were at a distance greater than 500 meters. Specific routes of traffic, such as routes of feed trucks or other company trucks, were not available and therefore not included in this analysis.

The connectivity test indicated that eight of 15 premises that directly linked to the connecting route were infected, compared to one of 50 of not connected premises that was infected (Table 2). The relative risk was 26.67 (3.62, 196.49).

**Table 2. 2 x 2 table denoting the connectivity of the connecting route to the infected and uninfected premises**

|                       | HP/LPAI  | No Detection | Total        |
|-----------------------|----------|--------------|--------------|
| Connected             | 8        | 7            | 15           |
| Not connected         | 1        | 49           | 50           |
| Total                 | 9        | 56           | 65           |
|                       | Estimate | Lower 95% CI | Upper 95% CI |
| Prevalence Odds Ratio | 56.00    | 6.05         | 518.01       |
| Relative Risk         | 26.67    | 3.62         | 196.49       |

The assessment of distance to the connecting route indicated that six of the eight premises that were within 500 meters from the common connecting route were infected when compared to three of 57 premises infected that were outside 500 meters (Table 3). The relative risk was 14.25 (4.41, 46.00).

**Table 3. 2 x 2 table denoting the effect of distance to infected and uninfected premises**

|                       | HP/LPAI  | No Detection | Total        |
|-----------------------|----------|--------------|--------------|
| Within 500m           | 6        | 2            | 8            |
| Outside 500m          | 3        | 54           | 57           |
| Total                 | 9        | 56           | 65           |
|                       | Estimate | Lower 95% CI | Upper 95% CI |
| Prevalence Odds Ratio | 54.00    | 7.47         | 390.35       |
| Relative Risk         | 14.25    | 4.41         | 46.00        |

These observations suggest that the common connecting route might be a key component in the observed north-south pattern of infected premises reported in this outbreak. The network of north-to-south roadways may have served as a point source for spreading the virus through movement of birds, personnel, feed, litter, or compost. These resources have been implicated in spread of LPAI in past outbreaks (Vieira et al., 2009; Halvorson, 2009).

One of the companies evaluated their movement records along this connecting route for the 30 days prior to HPAI detection and provided the following information. They evaluated supervisor movement and live bird-haul truck movement (both full and empty trucks). The only identified vehicles from those evaluated movements that travelled the entire connecting route were empty live-haul trucks. These trucks were disinfected with hot water wash prior to leaving the processing premises. Data for the movement of litter trucks and feed trucks was not available for evaluation, but the company noted that no common feed truck would have visited all the infected premises.

Field observations indicated premises on hilltops were more commonly infected compared to premises in a valley. Six of the 25 premises that were on hilltops were infected when compared to three of 40 premises that were in valleys that were infected (Table 4). The relative risk was 3.2 (0.87, 11.66). While this is indicative of an association, it does not account for location of the road, which also follows the slope and is a likely confounder in this analysis.

**Table 4. 2 x 2 table denoting the effect of hilltop to infected and uninfected premises**

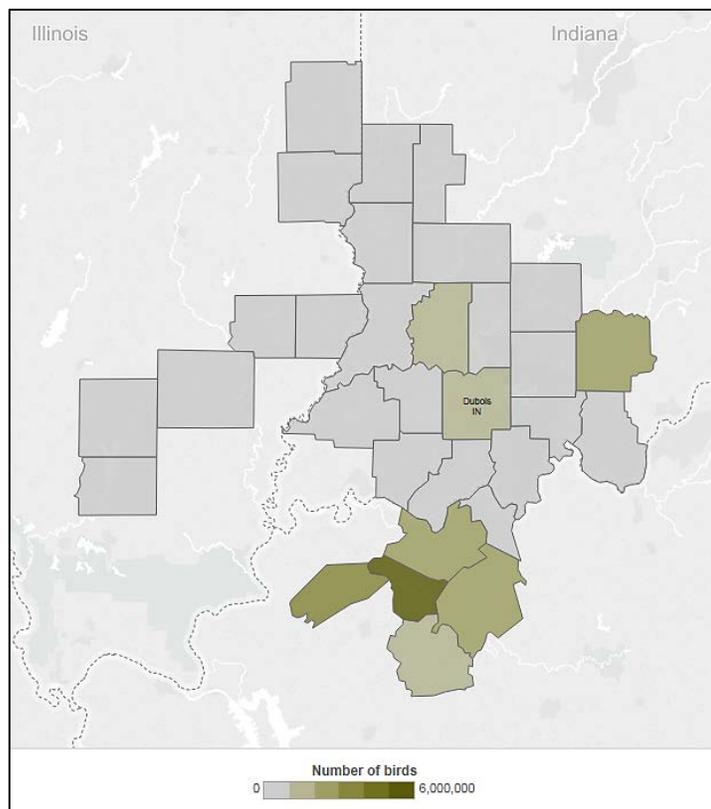
|                | HP/LPAI  | No Detection | Total     |
|----------------|----------|--------------|-----------|
| On hilltop     | 6        | 19           | 25        |
| Not on hilltop | 3        | 37           | 40        |
| <b>Total</b>   | <b>9</b> | <b>56</b>    | <b>65</b> |

|                       | Estimate | Lower 95% CI | Upper 95% CI |
|-----------------------|----------|--------------|--------------|
| Prevalence Odds Ratio | 3.89     | 0.88         | 17.32        |
| Relative Risk         | 3.20     | 0.88         | 11.66        |

**G. Population at Risk**

For HPAI/LPAI to enter a poultry population, the virus must enter the environment and have a susceptible population to infect. Dubois County has one of the higher densities of poultry compared to its neighboring counties (Figure 9).



**Figure 9. NASS 2012 turkey and chicken inventory of poultry by county.**

## H. Conclusions

This assessment describes the large-scale attributes present that may have influenced the introduction of the H7N8 virus and identifies a few hypotheses about these attributes, which could stimulate future studies. However, it is not possible to determine if wild birds persisted in this area due to environmental conditions, or if waterfowl behavior in late 2015 and early 2016 was normal but management practices influenced the introduction, or whether other factors led to the introduction of H7N8.

Several environmental factors present in Dubois County appeared to differ from neighboring counties (watershed connectivity, presence of a dense poultry population) or from previous years (higher temperature and precipitation); however, a statistical analysis was not completed at this time. While temperature and precipitation in the counties examined did not appear different from Dubois, those conditions may have been favorable for migratory waterfowl to remain longer in Indiana. The presence of a large mass of public land with accessible water may also have attracted waterfowl, although this area of Indiana may be where waterfowl normally congregate during migration.

If infected waterfowl were present, Dubois County is downstream and within the same watershed of Hoosier National Forest; therefore, birds or their feces may have moved into the county. Neighboring counties downstream did not have as sizeable of a poultry population to become infected.

Results from the road assessment indicate that a premises' proximity to the identified common route may have been a risk factor for developing infection, yet no specific vehicle movement or other variable can be attributed to that risk at this time.

## I. Next Steps

Additional analysis of environmental conditions in previous LPAI outbreaks would help determine if the environmental conditions present in Dubois County were a contributing factor(s) to the presence of infected waterfowl and subsequent poultry infection. Given the likely association between the north-south connecting route, it may be valuable to investigate more specific vehicle movement along this common route to analyze whether an association can be made among resource movement patterns and infected premises.

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## IV. PHYLOGENETIC ANALYSIS AND DIAGNOSTICS - UPDATED

### A. North American H7N8 Viruses

This report describes the H7N8 HPAI virus from a commercial turkey flock in Dubois County, Indiana, confirmed by the National Veterinary Services Laboratories (NVSL) in January 2016.<sup>1</sup> The NVSL confirmed H7N8 HPAI in the index flock by cleavage site analysis from partial hemagglutinin (HA) gene sequence obtained directly from the sample. Eight subsequent H7N8 detections from control zone surveillance were confirmed to be LPAI with high similarity to the HPAI virus excluding the insertion at the cleavage site in the HPAI virus. Due to the potential risk of mutation, control actions appropriate to HPAI were followed. There have been no further detections since January 16.

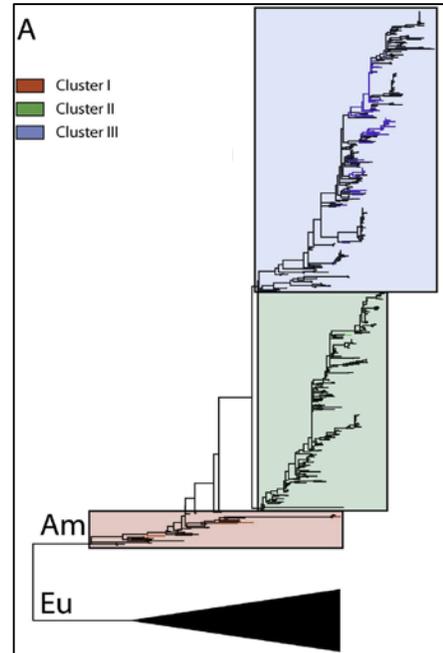
This is the first detection of an H7N8 HPAI virus. The H7N8 viruses are not related to the Eurasian H5 HPAI viruses from 2014-15. Based upon the full genome sequence<sup>2</sup> of eight H7N8 viruses representing seven different premises, the index case of H7N8 HPAI and subsequent detection of H7N8 LPAI are of North American wild bird lineage with high similarity to other recent wild bird viruses (North American Cluster III viruses; blue box in Figure 10<sup>3</sup>). They are highly similar to each other across all eight genes (excluding the multi-basic amino acid insertion at the cleavage site responsible for the mutation to HPAI). The H7N8 HPAI and LPAI viruses are also highly similar across six of eight gene segments to a recent wild bird detection of H7N8 LPAI in Kentucky (lesser scaup collected November 28, 2015). This suggests that reassortment, which is common to influenza A viruses in wild birds, occurred prior to the virus being introduced in turkeys in Indiana. However, the mutation to the highly pathogenic form identified at a single farm likely occurred during replication of the virus in poultry.

**Figure 10. Schematic phylogenetic tree of the HA1 nucleotide sequences of H7 AIVs.**

The tree was constructed based on the phylogeny inferred by using the maximum-likelihood method. Boxes represent the 3 major genetic clusters; the Eurasian lineage (EU) is represented by the large black triangle. Am = North American lineage.

The H7N8 viruses are located in Cluster III.

*Courtesy of Xi-Feng Wan et al, Department of Basic Sciences, College of Veterinary Medicine, Mississippi State University.*



<sup>1</sup> Virus availability announcement:

[https://www.aphis.usda.gov/animal\\_health/vet\\_biologics/publications/notice\\_16\\_02.pdf](https://www.aphis.usda.gov/animal_health/vet_biologics/publications/notice_16_02.pdf)

<sup>2</sup> Genbank IDs: H7N8 HPAI KU558903-8910; H7N8 LPAI KU585905-12 and KU585913-20

<sup>3</sup> Xu Y, Bailey E, Spackman E, et al. 2016. Contemporary H7 Avian-Origin Influenza A Viruses from North America (Scientific Reports, 6: 20688)

Additional genetic analysis of the eight H7N8 viruses for which whole genome sequence was obtained suggests a point source introduction followed by lateral/secondary spread, and provides potential insight into virus evolution and the epidemiology of this outbreak.

In general, influenza A viruses, including H5 and H7 subtypes, circulate in wild waterfowl in a form that is low pathogenic for chickens, and to date H7 HPAI viruses have been documented after the virus has circulated in poultry. The only H5s recognized to circulate in wild birds in a form that is highly pathogenic for chickens are the goose Guangdong-lineage viruses, such as those involved in the Eurasian H5 clade 2.3.4.4 outbreak in 2014-15 (Table 5). North American H7N8 LPAI virus has been previously detected in wild bird surveillance in the United States, but this is the first instance of H7N8 HPAI virus detection in any species.

All H7N8 viruses detected from this event are of North American wild bird lineage, the HPAI and LPAI viruses are highly similar across all eight genes, excluding the multibasic amino acid insertion at the cleavage site in the HPAI virus, and genetic analysis suggests a point source introduction followed by lateral/secondary spread.

**Table 5. Major differences between 2014-15 Eurasian H5 viruses and the North American H7N8 influenza A viruses (IAV).**

|  | Eurasian (EA) H5N8   | EA/AM H5N2 reassortant | North American (AM) H7N8  |
|--|--|------------------------|---|
| Related to Asian H5N8?                                     | Yes<br>(all 8 genes)   | Yes<br>(5 genes)       | No<br>(not even N8)   |
| Related to North American IAV?                             | No   | Yes<br>(3 genes)       | Yes<br>(all 8 genes)  |
| Was the initial virus adapted to poultry?                  | No   | No                     | Unlikely - <i>research in progress</i>  |
| Were virus adaptations noted after circulation in poultry? | Yes (CA)   | Yes (Midwest)          | Yes (LPAI>HPAI)   |
| Species adaptation/ virus infectivity                      | The initial viruses detected in the U.S. were poorly adapted to poultry with low infectivity and had prolonged mean death times; unlike previous Asian H5N1 viruses, the Eurasian H5 viruses may initially circulate undetected  |                        | <i>Research in progress</i> – the H7N8 is a wild bird lineage virus, and like other LPAI viruses may circulate undetected |
| Reservoir  | The Asian H5 lineage (goose Guangdong 1996) are the <u>only</u> HPAI viruses recognized to circulate in wild waterfowl and may be the result of spillover from domestic poultry into wild birds in Asia - <i>Prior to 1996, no HPAI viruses were recognized to circulate in wild birds</i> |                        | Wild waterfowl are the natural reservoir for influenza A viruses (H1-H16)   |
| Mode of spread   | Molecular/epi data supports point source introduction and lateral/secondary spread occurring at the same time (esp. early in Midwest turkeys); outbreaks at the end of the epi curve largely attributed to lateral/secondary spread  |                        | Molecular data supports point source introduction with subsequent lateral/secondary spread                                |
| Control  | No difference - stamp out HPAI   |                        |   |

## **B. Public Health Aspects**

No cases of H7N8 HPAI virus infection have been reported in humans at this time, and no human infections associated with avian influenza A viruses of this particular subtype (i.e., H7N8) have ever been reported.

All viruses to date lack key amino acid substitutions associated with human-like receptor binding or substitutions in the polymerase or other internal genes associated with increased virulence and transmission in mammals.

No known markers of neuraminidase inhibitor (Oseltamivir) resistance have been identified.

Efforts to monitor the health of response workers and on-farm personnel continue.

## **C. Poultry Vaccine Strain Selection Considerations**

Genetic, antigenic, and growth characteristics are considered for selection of poultry candidate strains. Experimental studies in poultry indicate that antibody to the neuraminidase protein does not play a significant role in protection. Antigenic characteristics and challenge studies will be used to evaluate protection of existing and candidate vaccines; ongoing evaluation of viruses for antigenic drift will continue.

## **D. Diagnostics and Characterization for Influenza A viruses**

Molecular diagnostic tests for influenza A virus (IAV) are used across the National Animal Health Laboratory Network (NAHLN) in the United States. As primary surveillance tools, the NAHLN H5 and H7 assays are broadly reactive, and do not distinguish geographic lineage or pathotype. There were no issues with detection of the H7N8 viruses from Indiana using the NAHLN assays. The NVSL uses Sanger sequencing protocols to generate partial HA/NA sequence directly from the sample for confirmation, pathotyping, and subtype determination where sufficient virus is present. Whole genome sequencing is conducted on all isolated viruses and select viruses are further characterized by pathotype assay in specific pathogen-free chickens.

USDA's NVSL collaborates with the Southeast Poultry Research Laboratory (SEPRL), the Influenza Division of the Centers for Disease Control and Prevention (CDC), and other key partners to rapidly share genetic and biological materials. Consensus data from whole genome sequencing is used to monitor the virus evolution and assess risk to veterinary or public health based upon presence/absence of specific amino acid substitutions or protein motifs. Analysis of sequence data includes phylogeny of all eight segments and determination of amino acid substitutions across the HA1 protein.

## **V. CASE-CONTROL STUDY TO INVESTIGATE H7N8 VIRUS IN TURKEYS IN INDIANA - UPDATED**

### **A. Background**

USDA-APHIS conducted a case-control study among commercial turkey operations in Indiana to investigate potential risk factors for infection with H7N8 avian influenza virus. We conducted in-person interviews with producers on nine case farms where H7N8 was detected (HPAI, n=1; LPAI, n=8) and 30 control farms located within the control area. Respondents were instructed to answer questions for the 2-week time period from Jan. 1 to Jan. 14, 2016. The questionnaire addressed general premises characteristics, biosecurity practices, wild birds, employees and visitors, vehicles and equipment, litter handling, and dead bird disposal (see [appendix](#)). In addition, questions were asked about one infected barn (case farms) or one barn containing birds during the 2-week time period (control farms). Data were entered into a SAS data set. Statistical comparisons between case and control farms, including potential confounding influence of one variable upon another, were not attempted because the number of case farms was small. The percentage of case farms and control farms having farm-level (Table 6) and barn-level (Table 7) characteristics for selected variables are reported here. Results for the complete set of variables are included in the [appendix](#).

### **B. Description of Study**

Although case and control farms were similar in size (total number of birds), control farms had more employees than case farms and control barns held more birds than case barns. Birds on case farms were older than birds on control farms. The percentage of control barns with young birds may have been somewhat biased due to selling off birds close to market age. However, none of the case barns had birds younger than 8 weeks of age (as of Jan. 14, 2016) compared to nearly half of control barns. Additionally, one-third of case farms versus two-thirds of control farms reported that they raised brooder birds on the farm. Temperatures in control barns were higher than in case barns, which may be related to having younger birds.

Over half of case farms and about one-fourth of control farms were located east of a public road. Case barns and control barns were located a similar distance from the nearest road, but case barns were located closer than control barns to dead bird disposal and litter compost areas. Being near a dead bird disposal area was found to be a risk factor in a previous HPAI case-control study on table egg layer farms (USDA 2015). All case farms and three-fourths of control farms had any water bodies located within 350 yards of the farm. Ponds were located within 350 yards of all case farms and 60 percent of control farms. Waterfowl and other birds were rarely observed on the farm, in nearby fields, or on water bodies during the 14 days prior to detection of infection on either case or control farms (see [appendix](#)). Wild mammals were seen more frequently on case farms. Also, poultry feed was accessible to wild birds, wild animals, and rodents on a higher percentage of case farms than control farms. Presence of wild mammals was found to be a risk factor in a previous LPAI (H7) case-control study in Virginia (McQuiston et al., 2005).

A higher percentage of case farms than control farms had any visitors enter a barn; in particular, veterinarians, company service persons, and occasional workers. A higher percentage of case barns than control barns were dealing with another health concern in the flock, so it is possible the veterinarian or service person visit may have been related to this problem. Company service person visit was found to be a risk factor in the previous HPAI case-control study on table egg layer farms.

One-third of case farms had a hard surface entry pad that was cleaned and disinfected, compared to two-thirds of control farms. This practice was found to be a protective factor in the previous HPAI case-control study on table egg layer farms.

**Table 6. Farm-level characteristics**

| Characteristic  | Percent case farms    |                          |                           | Percent control farms |                          |                           |
|---|-----------------------|--------------------------|---------------------------|-----------------------|--------------------------|---------------------------|
| Farm size (number of birds on farm on reference date):  |                       |                          |                           |                       |                          |                           |
| Less than 20,000  | 44                    |                          |                           | 55                    |                          |                           |
| 20,000 or more  | 56                    |                          |                           | 45                    |                          |                           |
| Stage of production:  |                       |                          |                           |                       |                          |                           |
| Brooder   | 33                    |                          |                           | 62                    |                          |                           |
| Grower  | 100                   |                          |                           | 93                    |                          |                           |
| Breeders  | 0                     |                          |                           | 0                     |                          |                           |
| Other   | 0                     |                          |                           | 0                     |                          |                           |
| If both grower and brooder:   |                       |                          |                           |                       |                          |                           |
| In different barns  | 100                   |                          |                           | 100                   |                          |                           |
| Sex of market type:   |                       |                          |                           |                       |                          |                           |
| Hens (HH or LH)   | 0                     |                          |                           | 10                    |                          |                           |
| Toms (FP)   | 100                   |                          |                           | 87                    |                          |                           |
| Both hens and toms  | 0                     |                          |                           | 3                     |                          |                           |
| Any water bodies within 350 yards:  | 100                   |                          |                           | 77                    |                          |                           |
| Pond  | 100                   |                          |                           | 60                    |                          |                           |
| Direction from public road to farm:   |                       |                          |                           |                       |                          |                           |
| East/NE/SE  | 56                    |                          |                           | 27                    |                          |                           |
| Wild mammals (or evidence) observed in or around poultry barns during the 14-day defined risk period              | 88                    |                          |                           | 41                    |                          |                           |
| Access to poultry feed:   |                       |                          |                           |                       |                          |                           |
| Wild birds  | 38                    |                          |                           | 10                    |                          |                           |
| Wild animals  | 38                    |                          |                           | 10                    |                          |                           |
| Rodents   | 25                    |                          |                           | 3                     |                          |                           |
| Number of employees:  |                       |                          |                           |                       |                          |                           |
| 1   | 38                    |                          |                           | 7                     |                          |                           |
| 2   | 25                    |                          |                           | 29                    |                          |                           |
| 3 or more   | 38                    |                          |                           | 64                    |                          |                           |
| Visitor type during the 14-day risk period:   | Visitors <sup>1</sup> | Avg # times <sup>2</sup> | Entered barn <sup>3</sup> | Visitors <sup>1</sup> | Avg # times <sup>2</sup> | Entered barn <sup>3</sup> |
| Private or company veterinarian   | 13                    | 2                        | 25                        | 0                     |                          |                           |
| Company service person  | 88                    | 2                        | 88                        | 72                    | 2                        | 57                        |
| Catch crew (bird removal)   | 25                    | 1                        | 14                        | 7                     | 1                        | 10                        |
| Feed delivery personnel   | 88                    | 8                        | 13                        | 97                    | 4                        | 8                         |
| Occasional worker (e.g., family member, part-time help over holiday)  | 50                    | 8                        | 38                        | 23                    | 2                        | 20                        |
| Any visitors enter the barn   | 100                   |                          |                           | 57                    |                          |                           |
| <sup>1</sup> Percent of all farms that had the visitor  |                       |                          |                           |                       |                          |                           |
| <sup>2</sup> For farms with the visitor   |                       |                          |                           |                       |                          |                           |
| <sup>3</sup> Percent of farms where the visitor entered the barn, out of all farms (with and without the visitor) |                       |                          |                           |                       |                          |                           |

**Table 7. Barn-level characteristics**

| Characteristic   | Percent case barns | Percent control barns |
|--|--------------------|-----------------------|
| Number of birds placed in this barn:   |                    |                       |
| Fewer than 4,000   | 0                  | 4                     |
| 4,000 to 9,999   | 100                | 64                    |
| 10,000 or more   | 0                  | 32                    |
| Type(s) of poultry are present in this barn:   |                    |                       |
| Brooder  | 0                  | 26                    |
| Grower toms  | 100                | 74                    |
| Grower hens  | 0                  | 4                     |
| Breeders   | 0                  | 0                     |
| Other  | 0                  | 0                     |
| Age of birds (weeks):  |                    |                       |
| Less than 8  | 0                  | 44                    |
| 8 to less than 14  | 25                 | 15                    |
| 14 or more   | 75                 | 41                    |
| Another health concern in this flock during the defined risk period                        | 44                 | 11                    |
| Hard-surface entry pad (e.g., concrete, asphalt):  |                    |                       |
| Cleaned and disinfected  | 33                 | 68                    |
| Cleaned only   | 11                 | 14                    |
| Not cleaned or disinfected   | 33                 | 7                     |
| No hard-surface entry pad  | 22                 | 11                    |
| Types of people who entered this barn during the 14-day defined risk period:               |                    |                       |
| Private or company veterinarian  | 29                 | 4                     |
| Company service person   | 88                 | 61                    |
| Occasional worker (e.g., family member, part time help over holiday)                       | 38                 | 21                    |
| Any visitor enter the barn   | 100                | 68                    |
| Median minimum and maximum temperatures in the barn during the 14-day defined risk period? |                    |                       |
| Minimum  | 55                 | 61                    |
| Maximum  | 65                 | 69                    |
| Median distance this barn (in yards) from:   |                    |                       |
| Dead bird disposal/holding area including carcass bin for rendering                        | 59                 | 100                   |
| Litter compost (for barns where litter is composted)                                       | 50                 | 100                   |
| Nearest road   | 100                | 100                   |

### **C. References**

- USDA–APHIS. Epidemiologic and other analyses of HPAI-affected poultry flocks: September 9, 2015. Appendix B. HPAI Case Control Questionnaire – Layers. Available at [https://www.aphis.usda.gov/animal\\_health/animal\\_dis\\_spec/poultry/downloads/Epidemiologic-Analysis-Sept-2015.pdf](https://www.aphis.usda.gov/animal_health/animal_dis_spec/poultry/downloads/Epidemiologic-Analysis-Sept-2015.pdf)
- McQuiston, J. H., L. P. Garber, B. A. Porter-Spalding, J. W. Hahn, F. W. Pierson, S. H. Wainwright, D. A. Senne, T. J. Brignole, B. L. Akey, and T. J. Holt, 2005. Evaluation of risk factors for the spread of low pathogenicity H7N2 avian influenza virus among commercial poultry farm. *J. Am. Vet. Med. Assoc.* 226:767–772. 2005.

## VI. PRELIMINARY ASSESSMENT OF HPAI 2016 MORTALITY SHEET DATA - NEW

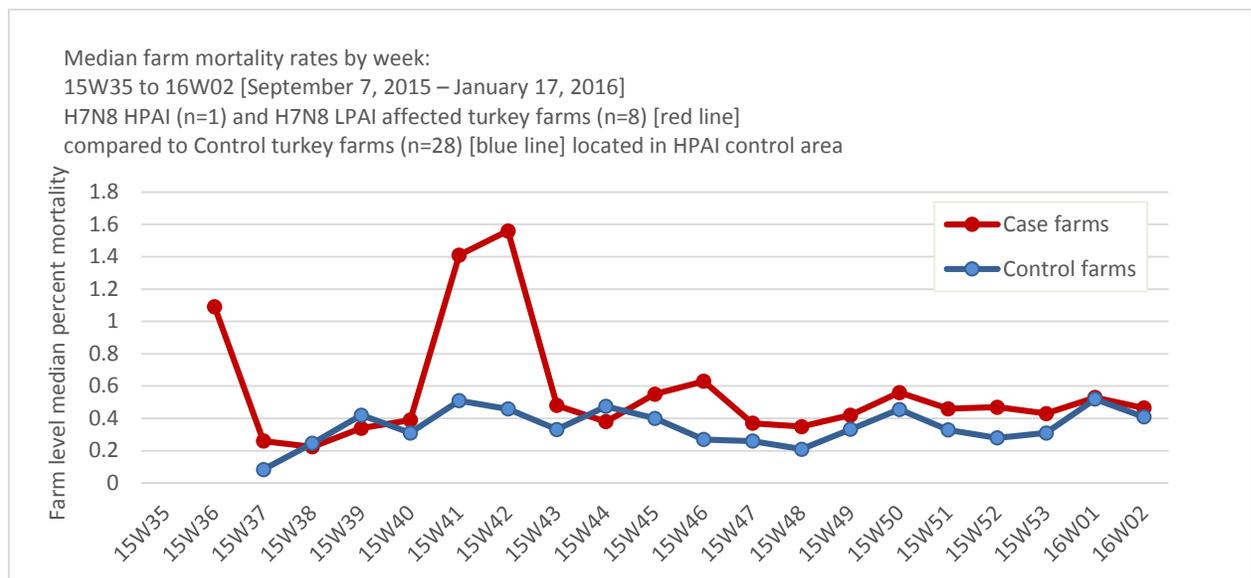
Data from daily mortality sheets recorded in individual turkey barns were submitted for analysis from one H7N8 HPAI-affected farm (with four barns) and eight H7N8 LPAI-affected farms (with 26 barns). These farms and barns were detected during the 2016 HPAI outbreak in Indiana. In addition, data were collected from 28\* control farms (77 barns) during a similar period in the HPAI control area.

The daily mortality data were used to obtain weekly mortality rates per barn, which were combined to obtain average weekly mortality rates per farm for case farms as well as control farms. High mortality was observed during the first 2 weeks after placement in many of the barns. This mortality was not likely associated with avian influenza. Consequently, the first 2 weeks of mortality data for each barn, post placement, were excluded from further analyses.

The mean average mortality rate by week of the case farms and control farms was calculated to compare the mortality rates over the time frame of weeks 15W37 through 16W02 (September 7, 2015 – January 17, 2016) to look for potential case farm risk weeks identified by the mortality rates.

### A. Findings

The median averaged farm mortality rates for case farms were compared to control farms. The pattern of averaged farm mortality rates between the case farms and control farms did not differ dramatically, except during the weeks of 15W41 (October 5 – 11, 2015) and 15W42 (October 12 – 18, 2015), with case farms demonstrating an average mortality rate that was 0.9 percent and 1.1 percent higher, respectively, for those weeks. Also, during the week of 15W46 (November 9 – 15, 2015), case farms demonstrated an average mortality rate that was 0.36 percent higher for that week (Figure 11).



**Figure 11. Median farm mortality rates by week**

Seven of the eight H7N8 LPAI-affected farms noted a gradual slight increase in mortality rates during the last 1 to 2 weeks of the time period when turkeys were present on the farms (i.e., from approximately January 11 through January 15, 2016). Generally, however, mortality remained below 0.3 percent during that time. On the H7N8 HPAI-affected farm, the unaffected barns showed a

similar pattern as the H7N8 LPAI farms during the last 1 to 2 weeks, whereas the HPAI-affected barn itself peaked on January 15, 2016, at 19.20 percent mortality. A number of control farms/barns also exhibited an apparent pattern of increasing mortality rates during weeks of December 2015 and January 2016 (22/28, 75 percent).

## **B. Discussion**

This preliminary comparison of mortality patterns did not adjust for age of the birds in the barns. Further analysis is needed to account for differences in mortality that may have occurred related to the age of birds. The spike in mortality of the HPAI-infected farm did trigger a response. APHIS emergency management plans state that investigations should be conducted on daily mortality exceeding 0.1 percent or weekly mortality exceeding 1 percent<sup>4</sup>. In “tom” flocks, mortality gradually increases after 16 weeks and may reach 1 percent per week by the time they are processed, which was seen in the case and control flocks. Extremely high daily mortality (>1/100) constitutes an emergency and should be reported and investigated to rule out the possibility of a foreign animal disease.

At this time, the importance of the mortality differences seen in October and November is uncertain. Although the relationship between this spike in mortality and LPAI is unknown, it may have been a predisposing factor. This mortality pattern difference between LPAI infected and uninfected premises was unexpected, especially in comparison to similar mortality patterns, as was noted, beginning November 16, 2015.

## **C. Next steps**

1. APHIS plans to conduct more detailed analyses, including age-adjusted analyses of the weekly mortality rates by barns.
2. Water consumption per barn will be evaluated, comparing consumption in barns on case farms to control farms.

\*For this preliminary review, submissions of mortality sheets were interpretable for use from 28/30 control farms.

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<sup>4</sup> Emergency Management Plans  
[[https://www.aphis.usda.gov/animal\\_health/emergency\\_management/.../poultry\\_ind\\_manual.pdf](https://www.aphis.usda.gov/animal_health/emergency_management/.../poultry_ind_manual.pdf)]

## VII. ON-FARM SAMPLING: PRELIMINARY REPORT - UPDATED

### Sampling for Influenza A Virus in Synanthropic Wildlife at Infected Premises

#### A. Objective

To evaluate the potential for synanthropic wildlife associated with commercial turkey flocks to become exposed or infected with highly pathogenic or low pathogenic influenza A (H7N8) viruses, USDA-APHIS Wildlife Services sampled peri-domestic birds and mammals on farms that had been infected with an H7N8 influenza A virus.

Four farms with confirmed H7N8 infections were investigated (Table 8). All flocks were located in southern Indiana. Sampling at confirmed infected sites was conducted within 2 weeks after viral excretion was confirmed in poultry. Three of the four infected flocks were depopulated prior to wildlife sampling and one of the flocks was being depopulated during sampling.

**Table 8. Summary of Infected Flocks**

| Site    | Approximate Flock Size | Virus             | Date H7N8 Confirmed by NVSL | Wildlife Sampling Period |
|---------|------------------------|-------------------|-----------------------------|--------------------------|
| DuBois8 | 36,695                 | Low Pathogenic    | 1/16/2016                   | 1/20-23/2016             |
| DuBois1 | 62,109                 | Highly Pathogenic | 1/14/2016                   | 1/25-27/2016             |
| DuBois9 | 16,591                 | Low Pathogenic    | 1/16/2016                   | 1/28-29/2016             |
| DuBois6 | 24,732                 | Low Pathogenic    | 1/16/2016                   | 1/30/2016                |

#### B. Sampling Procedures

We captured wild birds and wild mammals on farms, primarily around farm structures. Birds were captured using mist nets and baited funnel traps. Mammals were trapped using baited collapsible Sherman traps (mice) and baited Tomahawk traps. We also collected environmental samples in and around feed hoppers and barns.

Captured individuals were sampled to test for infection with influenza A viruses (IAV) by collecting swabs, washes, and tissues. Prior exposures were also investigated by testing serum. From birds, we collected an oral swab and a cloacal swab. From targeted avian species (e.g., European starlings), we also collected a blood sample and lung tissue. From mammals, we collected an oral swab, nasal swab/wash, and external swab. From targeted mammalian species, we also collected a blood sample and lung tissue sample. Swabs, washes, and tissue samples were placed in 1-3mL of viral transport media and stored chilled. Blood was collected into serum separator tubes, allowed to clot, and centrifuged. We shipped samples overnight on ice to testing laboratories within 24 hours during the week or stored them in a refrigerator and then shipped overnight on ice.

#### C. Laboratory Procedures

Swabs, washes, and tissue samples were screened for influenza A virus (IAV) matrix gene RNA via real-time reverse transcriptase polymerase chain reaction (RRT-PCR). The Avian Veterinary Diagnostic Laboratory at Colorado State University conducted matrix gene RRT-PCR testing of avian oral and cloacal swabs, while the National Wildlife Research Center Virology Laboratory conducted all other matrix gene RRT-PCR. Per the National Animal Health Laboratory Network (NAHLN) protocol, any cycle threshold (Ct) value >0 was considered positive for viral RNA. Samples with Ct>0 by matrix gene RRT-PCR were submitted to the USDA's NVSL in Ames, IA, for confirmatory testing. Confirmatory testing included subtype confirmation using H5 and H7 assays targeting American

lineage viruses. All serum samples were screened for antibodies to influenza A using the IDEXX AI Multi-S Screen Ab test, which is a multi-species blocking enzyme linked immunosorbent assay (ELISA) targeting an epitope of the nucleoprotein. All serum samples with S/N ratios <0.7 were submitted to NVSL for hemagglutinin inhibition (HI) assay testing using the H7N8 virus as the antigen.

#### D. Results

Across the four sampled farms, we collected 297 samples from 81 individuals (primarily starlings; Table 9). We sampled 77 individual birds across four species and four individual mammals across three species (Table 10). European starlings were the most commonly sampled species. We also collected 40 environmental samples, of which all but four were presumed to be from European starlings.

PCR testing for all oral and cloacal swabs, tissues, and environmental samples is complete and all samples were negative. ELISA testing for all serum samples has been completed. No samples were positive for antibodies to influenza A virus, but three samples were suspect positive (S/N ratio between 0.5-0.7). These samples were submitted to NVSL for H7 specific HI testing and were negative (Table 11).

**Table 9. Summary of Samples Collected**

| Sample Type     | Total Number Collected | Number Collected from Birds | Number Collected from Mammals |
|-----------------|------------------------|-----------------------------|-------------------------------|
| Serum           | 65                     | 62                          | 2                             |
| Oral Swab       | 81                     | 77                          | 4                             |
| Cloacal Swab    | 77                     | 77                          | --                            |
| Nasal Swab/Wash | 4                      | --                          | 4                             |
| External Swab   | 2                      | --                          | 2                             |
| Lung Tissue     | 67                     | 63                          | 3                             |
| Environmental   | 40                     | 38                          | 2                             |

**Table 10. Summary of Animals Sampled**

| Species                | Scientific Name               | Numbered Sampled |
|------------------------|-------------------------------|------------------|
| House mouse            | <i>Mus musculus</i>           | 2                |
| White-tailed deer      | <i>Odocoileus virginianus</i> | 1                |
| Raccoon                | <i>Procyon lotor</i>          | 1                |
| European starling      | <i>Sturnus vulgaris</i>       | 63               |
| Dark-eyed junco        | <i>Junco hyemalis</i>         | 8                |
| White-throated sparrow | <i>Zonotrichia albicollis</i> | 4                |
| Song sparrow           | <i>Melospiza melodia</i>      | 2                |

**Table 11. Summary of positive samples for avian serum samples tested by ELISA**

| Sample   | Species           | ELISA            | S/N Ratio | HI       |
|----------|-------------------|------------------|-----------|----------|
| IDa01020 | European starling | Suspect Positive | 0.66      | Negative |
| IDa01026 |                   | Suspect Positive | 0.68      | Negative |
| IDa01027 | European starling | Suspect Positive | 0.61      | Negative |

## **E. Summary**

No wildlife samples from animals associated with infected premises showed evidence of influenza infection or exposure. However, several important observations were noted among the sampled farms. First, the high levels of ammonia associated with depopulation and composting likely lowered mouse populations in turkey barns and undoubtedly influenced the number of mice captured. Second, weather patterns appeared to influence bird capture success, as the bulk of the starlings sampled in this survey were captured when temperatures were unseasonably low and snow was on the ground. Capture success was greatly reduced at subsequent farms when snow was largely absent and European starlings moved to cornfields. Third, spilled feed from grain hoppers appeared to be a key factor attracting wildlife to the single farm that had multiple feed spills due to feed hopper tubing breaches. Fourth, forest birds made up the majority of wildlife observed in proximity to the farms but they did not spend time in close proximity to barns. Fifth, relatively low numbers of wildlife captures were expected during the winter months due to altered ecology during that season resulting in differing resource use of farms (wildlife farm visits may be sporadic, especially for birds), annual mortality (many animal populations are lowest during the winter), reduced resources, lack of breeding, and minimal movements.

It is clear that these factors contribute to the episodic use of poultry farms by wildlife, especially during the winter months. This periodic use of farms by wildlife undoubtedly contributed to low capture rates. As such, considering that the prevalence of IAV in the natural wildlife reservoir (i.e., waterfowl and aquatic birds) is relatively low during most time frames, it is a reasonable assumption that the natural prevalence in bridge species (synanthropic avian and mammalian species such as European starlings, mice, and skunks) is even lower. Therefore, detection of infection in these species is a challenging task even when the animals are relatively common on farmsteads. Although no positive wildlife species were detected within close proximity to farms in Indiana, the potential role of wildlife in the IAV case clusters of DuBois County should not be discounted. For example, following the HPAI outbreaks in Iowa, a PCR-positive European starling (lung tissue) and several avian species that were antibody-positive for HP H5N2 AIV were discovered. However, the avian populations at the Iowa sites, in general, were more diverse and more stable when compared to those sampled in Indiana.

Anecdotally, several owners of the sampled premises reported the presence of large numbers of European starlings and/or Canada geese (on some farms) prior to the outbreaks. Consequently, we plan to experimentally inoculate starlings and possibly geese with the H7N8 viruses to determine if these species can host these strains and shed viruses at levels that are epidemiologically significant.

This effort highlights the need to survey multiple poultry facilities in several regions of the United States to assess wildlife interactions with poultry facilities. Similar studies in Canada have been used to inform risk assessments and to develop lists of key species of concern. A comparable approach in the United States could be used to develop lists of key wildlife species of concern to the poultry industry. Once developed, a species list could guide experimental infection studies to assess the reservoir competence of select synanthropic wildlife species.

## **F. Acknowledgements**

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## VIII. LITERATURE REVIEW

### A. Latent and Infectious Periods for H7 HPAI Virus Strains

#### Background

The following is a brief summary of the latent and infectious period data for chickens and turkeys from literature review as well as unpublished data shared by the Southeast Poultry Research Laboratory in Athens, GA. The statistical distributions estimated from these data are useful for disease spread modelling and for evaluating the impact of various surveillance and movement protocols. Many of the analyses conducted to support emergency response and business continuity measures have been based on the latent and infectious periods for H5 HPAI viruses. The purpose of this review was to evaluate whether the latent and infectious periods for chickens and turkeys infected with H7 HPAI viruses differed greatly from what has been seen with H5 HPAI viruses.

#### Bird-level infectious period and mean time to death for H7 HPAI

There is considerable variability in the estimated infectious period for different H7 HPAI strains. The mean infectious period estimate was 6.3 days for Netherlands H7N7 HPAI in chickens (Table 12). Given the available data, only the mean time to death could be estimated for the Jalisco H7N3 HPAI strain. The mean time to death in chickens for this strain was around 2.3 days and suggests a shorter infectious period compared to the Netherlands strain.

In turkeys, the estimated infectious period was 6.2 days for the Netherlands H7N7 HPAI strain and 1.47 days for the Italy H7N1 HPAI strain. The mean time to death for the Mexico H7N3 HPAI strain in turkeys was 2.47 days.

**Table 12. Summary of infectious period and time to death data for H7 HPAI strains**

| HPAI strain           | Source  | Species  | Parameter estimated | Estimated value / Distribution Days               |
|-----------------------|---|----------|---------------------|---|
| H7N3 HPAI Mexico      | Unpublished data (Erica Spackman)                               | Turkeys  | Time to death       | 2.47 days (95% C.I., 0.9-4.92). Gamma (5.96,0.41) |
| H7N3 HPAI Mexico      | Unpublished data. Kapczynski (personal communication)           | Chickens | Time to death       | 1.9 (95% C.I., 0.9-4.92), Gamma (10.7,,18)        |
| H7N3 HPAI Mexico      | Bertran 2013; Kapczynski et al 2013; Spackman et al 2014. [1-3] | Chickens | Time to death       | 2.25; 2.3   |
| H7N7 HPAI Netherlands | Vandergoot et al 2005 [4]                                       | Chickens | Infectious period   | 6.3(95% C.I., 3.9-8.7)                            |
| H7N7 HPAI Netherlands | Bos et al 2008 [5]  | Turkeys  | Infectious period   | 6.2 days (range 5-8)                              |
| H7N7 HPAI Netherlands | White paper- Templeton et al, Maas et al 2009 [6]               | Chicken  | Time to death       | 5.1 (95% C.I. 3.5-6.9);                           |
| H7N7 HPAI Netherlands | CEAH analysis [4, 6]  | Chicken  | Infectious period   | 4.1 (95% C.I. 2.3-5.6); Gamma (5.738, 5.445)      |
| H7N1 HPAI Italy       | Saenz et al 2012 [7]  | Turkeys  | Infectious period   | 1.47 (95% C.I., .3-3); Gamma (2.199,1.668)        |

### Bird-level latently infected period

The estimated mean latent period in turkeys was 0.8 days for Netherlands HPAI H7N7 and 0.4 days for Italy H7N1 HPAI strain (Table 13).

For chickens, the available experimental data was censored with testing for shedding at daily intervals. However, for both the Mexico H7N3 HPAI strain and Netherlands H7N1 HPAI strains, the inoculated chicken were shedding by the first day indicating a short latent period.

**Table 13. Summary of bird-level latent period data for H7 HPAI strains**

| HPAI strain           | Source                                    | Species  |
|-----------------------|---|--|
| H7N3 HPAI Mexico      | Kapczynski et al (personal communication) | Between 0-1 days: 5 chickens inoculated, all were shedding within 24 hours;                    |
| H7N7 HPAI Netherlands | Vandergoot et al 2005 [4]                 | Between 0-1 days: 10 chickens inoculated, all were shedding within 24 hours;                   |
| H7N7 HPAI Netherlands | Bos et al 2008 [5]                        | 0.7 days: 8/10 turkeys were shedding by 1 day and 2/10 turkeys by 2 days. Gamma (17.08, 0.04)  |
| H7N1 HPAI Italy       | Saenz et al., 2012 [7]                    | Mean 0.40 days: based on 10 inoculated turkeys. Suggested gamma distribution (Gamma 2.2,0.186) |

### Summary and implications for current outbreak

The infectious and latent period distribution data was summarized for three H7 HPAI strains. Overall, the infectious period was short (1.5-2.5 days) for H7N3 HPAI and H7N1 HPAI strains compared to Netherlands H7N7 HPAI strain, which had an infectious period around 6 days. The data indicate a short latent period (less than 1 day) for all the three strains summarized. These results are similar to the infectious and latent period distributions identified during the 2015 outbreak of HPAI in the United States.

These results suggest that the assumptions related to bird-level viral transmission dynamics used for modeling work and analysis based on the U.S. HPAI outbreak in 2015 are still appropriate for an outbreak involving a highly pathogenic H7 virus.

## B. Predominant Clinical Signs in Turkeys for Recent Outbreaks of H5 or H7 HPAI virus strains

### Background

The presence and severity of clinical signs of HPAI infection depend on the type of bird species affected. Infected wild and domestic ducks may be asymptomatic, whereas clinical signs in gallinaceous poultry are usually severe, resulting in high mortality. In poultry (chickens and turkeys), the clinical signs associated with HPAI infection include marked depression with ruffled feathers, lack of appetite, excessive thirst, decreased egg production, soft-shelled or misshaped eggs, respiratory signs (coughing and sneezing), watery diarrhea, or sudden, unexpected death. In turkeys, a cessation in flock vocalization (Cathedral Syndrome) often accompanies infection. Mature chickens frequently have swollen, cyanotic combs and wattles and edema surrounding the eyes.

The mortality rate in an infected flock can reach 100 percent. In mature birds, gross lesions on necropsy may consist of subcutaneous edema of the head and neck; fluid in the nares, oral cavity, and trachea; congested conjunctivae and kidneys; and petechial hemorrhages that cover the abdominal fat, serosal surfaces, peritoneum, and surface under the keel. In layers, the ovary may be

hemorrhagic or degenerated and necrotic. The peritoneal cavity is frequently filled with yolk from ruptured ova, causing severe airsacculitis and peritonitis in birds that survive longer than 7 days. In addition, necropsy of birds affected in the 1999–2001 H7N1 HPAI outbreak in Italy revealed pancreatitis in all species of birds; this was most pronounced in turkeys and chickens.[8]

### Clinical signs in turkeys

Clinical signs for HPAI infection in turkeys are different from those in chickens. Moreover, they vary depending on the HPAI virus strain (see Table 14). Respiratory signs may not be present in turkeys infected with HPAI, and producers should watch for other symptoms including neurological signs.

**Table 14. Summary of primary clinical signs seen in turkeys during recent outbreaks by HPAI strain**

| HPAI Strain | Clinical signs  | Source                            |
|-------------|---|-----------------------------------|
| H7N1        | Nervous signs   | Mutinelli et al., 2003 [8]        |
| H5N8        | Depression, loss of balance, drooping of wings, twitching   | McNulty et al., 1985 [9]          |
| H7N7        | Apathy, reduced vocalization, swollen sinuses, mucus from beak, lying down with neck extended                 | Elbers et al., 2004 [10]          |
| H5N1        | Attenuated motor functions, torticollis and nystagmus, general behavioral aberrations                         | Perkins and Swayne, 2001 [11]     |
| H5N1        | Reluctance to move, facial oedema, sinusitis, oculo-nasal discharge, and haemorrhages on the shanks           | Kilany et al., 2011 [12]          |
| H5N1        | Depressed, quiet flock, nearly all sitting down, apathy, fine head tremors                                    | Irvine et al., 2007 [13]          |
| H5N9        | Depressed, wings dropped, diarrhea, abnormal respiration was not seen, listless                               | Narayan, Lang and Rouse 1969 [14] |
| H5N2        | Lethargy, depressed, lack of vocalization neurological signs, occasional torticollis and blood from the mouth | 2015 US Outbreak Reports          |

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**APPENDIX A. FARM-LEVEL QUESTIONS - UPDATED**

| Characteristic   | Percent case farms | Percent control farms |
|--|--------------------|-----------------------|
| Farm size (number of birds on farm on reference date):                                     |                    |                       |
| Less than 20,000   | 44                 | 55                    |
| 20,000 or more   | 56                 | 45                    |
| Number of barns on the farm:   |                    |                       |
| 1 - 3  | 44                 | 67                    |
| 4 or more  | 56                 | 33                    |
| Marketing arrangement:   |                    |                       |
| Contract   | 100                | 97                    |
| Other  | 0                  | 3                     |
| Stage of production:   |                    |                       |
| Brooder  | 33                 | 62                    |
| Grower   | 100                | 93                    |
| Breeders   | 0                  | 0                     |
| Other  | 0                  | 0                     |
| If both grower and brooder:  |                    |                       |
| In barn at same time   | 0                  | 0                     |
| In same barn but at different times (e.g., placed as brooders and remain through grow-out) | 0                  | 0                     |
| In different barns   | 100                | 100                   |
| Birds have outside access  | 0                  | 3                     |
| Sex of market type:  |                    |                       |
| Hens (HH or LH)  | 0                  | 10                    |
| Toms (FP)  | 100                | 87                    |
| Both hens and toms   | 0                  | 3                     |
| Age of birds on farm:  |                    |                       |
| Multiple   | 22                 | 57                    |
| Single   | 78                 | 43                    |
| Other poultry on farm (e.g., broilers, layers, etc.)                                       | 0                  | 0                     |
| Certified Organic  | 0                  | 0                     |
| Enrolled in NPIP   | 100                | 97                    |
| Water bodies within 350 yards:   |                    |                       |
| Pond   | 100                | 60                    |
| Lake   | 0                  | 10                    |
| Stream   | 0                  | 31                    |
| River  | 11                 | 7                     |
| Wetland or swamp   | 0                  | 3                     |
| Wastewater lagoon  | 0                  | 3                     |
| Other  | 0                  | 7                     |

| Characteristic  | Percent case farms | Percent control farms |
|---|--------------------|-----------------------|
| Waterfowl seen on the above water bodies during the 14-day defined risk period: |                    |                       |
| Ducks   | 11                 | 7                     |
| Geese   | 0                  | 7                     |
| Shorebirds  | 22                 | 10                    |
| Other   | 0                  | 3                     |
| Distance (yds.) to the closest field where crops or hay are harvested:          |                    |                       |
| Less than 50  | 67                 | 57                    |
| 50 to 99  | 22                 | 17                    |
| 100 or more   | 11                 | 27                    |
| Crop grown in the above field:  |                    |                       |
| Corn  | 56                 | 48                    |
| Soybeans  | 22                 | 34                    |
| Alfalfa/grass   | 22                 | 17                    |
| Other   | 0                  | 0                     |
| Above field tilled last fall  | 33                 | 28                    |
| Above field actively worked during the 14-day defined risk period:              | 0                  | 3                     |
| Waterfowl seen on the field during the 14-day defined risk period:              | 0                  | 16                    |
| Other animals present on farm premises:   |                    |                       |
| Beef cattle   | 44                 | 40                    |
| Dairy cattle  | 0                  | 0                     |
| Horses  | 0                  | 10                    |
| Sheep   | 0                  | 3                     |
| Goats   | 0                  | 3                     |
| Pigs  | 0                  | 13                    |
| Dogs  | 33                 | 30                    |
| Cats  | 44                 | 30                    |
| Poultry or domestic waterfowl (noncommercial)                                   | 0                  | 3                     |
| Other   | 0                  | 7                     |
| Water source for poultry:   |                    |                       |
| Municipal   | 100                | 93                    |
| Well  | 11                 | 7                     |
| Surface   | 0                  | 0                     |
| Other   | 0                  | 0                     |
| Water treatments:   |                    |                       |
| Chlorination (excluding municipal)  | 0/1                | 2/2                   |
| Acidifiers  | 0                  | 14                    |
| Iodine  | 11                 | 60                    |
| Peroxide  | 11                 | 27                    |
| Other   | 0                  | 10                    |
| Windbreaks present on farm:   |                    |                       |

| Characteristic  | Percent case farms |                           |           |               | Percent control farms |                           |           |               |
|---|--------------------|---------------------------|-----------|---------------|-----------------------|---------------------------|-----------|---------------|
| Evergreen or juniper  | 0                  |                           |           |               | 0                     |                           |           |               |
| Deciduous trees   | 44                 |                           |           |               | 43                    |                           |           |               |
| Structural (e.g., hill, natural break)  | 11                 |                           |           |               | 20                    |                           |           |               |
| Distance to nearest public gravel or dirt road, excluding driveways (yds.):                                     |                    |                           |           |               |                       |                           |           |               |
| Less than 100   | 22                 |                           |           |               | 20                    |                           |           |               |
| 100 to 499  | 33                 |                           |           |               | 40                    |                           |           |               |
| 500 or more   | 44                 |                           |           |               | 40                    |                           |           |               |
| Direction from public road to farm:   |                    |                           |           |               |                       |                           |           |               |
| East/NE/SE  | 56                 |                           |           |               | 27                    |                           |           |               |
| House with people living in it on the property  | 44                 |                           |           |               | 37                    |                           |           |               |
| If yes, common drive entrance to farm and residence   | 100                |                           |           |               | 64                    |                           |           |               |
| More than one entrance to the farm that could provide access to the poultry area                                | 13                 |                           |           |               | 41                    |                           |           |               |
| Road surface on the farm that vehicles coming onto the operation drive on:                                      |                    |                           |           |               |                       |                           |           |               |
| Hard top/asphalt  | 0                  |                           |           |               | 21                    |                           |           |               |
| Gravel  | 89                 |                           |           |               | 76                    |                           |           |               |
| Dirt  | 0                  |                           |           |               | 0                     |                           |           |               |
| Other   | 11                 |                           |           |               | 3                     |                           |           |               |
| Access of the following vehicles:   | Perimeter only     | Enter farm/ not near barn | Near barn | Does not come | Perimeter only        | Enter farm/ not near barn | Near barn | Does not come |
| Garbage/dumpster pickup   | 11                 | 0                         | 11        | 78            | 7                     | 13                        | 7         | 73            |
| Propane delivery  | 11                 | 11                        | 22        | 56            | 0                     | 3                         | 83        | 13            |
| Feed delivery   | 0                  | 0                         | 100       | 0             | 0                     | 3                         | 97        | 0             |
| Renderer  | 0                  | 0                         | 0         | 100           | 0                     | 0                         | 0         | 100           |
| Company personnel   | 11                 | 0                         | 89        | 0             | 0                     | 7                         | 93        | 0             |
| Other business visitors   | 22                 | 11                        | 56        | 11            | 0                     | 0                         | 60        | 40            |
| Has gate to farm entrance   | 0                  |                           |           |               | 30                    |                           |           |               |
| Perimeter surrounded by security fence  | 0                  |                           |           |               | 7                     |                           |           |               |
| Vegetation mowed/bush hogged at least 3 times per month   | 100                |                           |           |               | 89                    |                           |           |               |
| Vehicle wash station/spray area used during the 14-day defined risk period                                      | 0                  |                           |           |               | 20                    |                           |           |               |
| Workers and visitors park in restricted area away from the poultry barns during the 14-day defined risk period: | Always             | Sometimes                 | Never     |               | Always                | Sometimes                 | Never     |               |
| Workers   | 78                 | 22                        | 0         |               | 13                    | 0                         |           | 87            |
| Visitors  | 33                 | 33                        | 33        |               | 7                     | 3                         |           | 90            |
| Rat/mouse bait stations used on the farm during the 14-day defined risk period                                  | 89                 |                           |           |               | 100                   |                           |           |               |

| Characteristic   | Percent case farms |                   |              | Percent control farms |                   |              |
|--|--------------------|-------------------|--------------|-----------------------|-------------------|--------------|
| If yes, frequency checked (times/month):   |                    |                   |              |                       |                   |              |
| Less often than 1  |                    | 14                |              |                       | 10                |              |
| 1  |                    | 71                |              |                       | 63                |              |
| 2 or more  |                    | 14                |              |                       | 27                |              |
| Frequency rodents observed during the 14-day defined risk period:                                    |                    |                   |              |                       |                   |              |
| Frequently (e.g., daily)   |                    | 13                |              |                       | 0                 |              |
| Occasionally (e.g., weekly)  |                    | 13                |              |                       | 31                |              |
| Never  |                    | 75                |              |                       | 69                |              |
| Beetle control used during the 14-day defined risk period:   |                    | 11                |              |                       | 47                |              |
| Beetles observed in poultry barns during the 14-day defined risk period:                             |                    | 50                |              |                       | 52                |              |
| Fly control (other than manure removal) used during the 14-day defined risk period                   |                    | 0                 |              |                       | 7                 |              |
| Intensity of flies observed in poultry barns during the 14-day defined risk period:                  |                    |                   |              |                       |                   |              |
| High   |                    | 0                 |              |                       | 3                 |              |
| Medium   |                    | 0                 |              |                       | 7                 |              |
| Low/none   |                    | 100               |              |                       | 90                |              |
| Wild mammals (or evidence) observed in or around poultry barns during the 14-day defined risk period |                    | 88                |              |                       | 41                |              |
| Access to poultry feed:  |                    |                   |              |                       |                   |              |
| Wild birds   |                    | 38                |              |                       | 10                |              |
| Wild animals   |                    | 38                |              |                       | 10                |              |
| Rodents  |                    | 25                |              |                       | 3                 |              |
| Pelleted feed  |                    | 100               |              |                       | 100               |              |
| Feed treatment:  |                    |                   |              |                       |                   |              |
| Formaldehyde   |                    | 0                 |              |                       | 0                 |              |
| Heat treated   |                    | 100               |              |                       | 100               |              |
| Within 100 yards of barns:   | <b>Often</b>       | <b>Some-times</b> | <b>Never</b> | <b>Often</b>          | <b>Some-times</b> | <b>Never</b> |
| Waterfowl  | 0                  | 25                | 75           | 3                     | 17                | 79           |
| Gulls  | 11                 | 0                 | 89           | 0                     | 0                 | 100          |
| Small perching birds   | 25                 | 50                | 25           | 48                    | 41                | 10           |
| Blackbirds/crows   | 13                 | 50                | 37           | 45                    | 34                | 21           |
| Other water birds  | 0                  | 13                | 87           | 0                     | 0                 | 100          |
| Wild turkeys, pheasants, quail   | 13                 | 13                | 75           | 0                     | 10                | 90           |
| Raptors  | 0                  | 38                | 62           | 3                     | 24                | 72           |
| Pigeons/doves  | 0                  | 0                 | 100          | 10                    | 21                | 69           |
| Other  | 0                  | 0                 | 100          | 0                     | 0                 | 100          |
| In the barn:   |                    |                   |              |                       |                   |              |
| Large birds (pigeons, crows)   | 0                  | 0                 | 100          | 0                     | 0                 | 100          |

| Characteristic   | Percent case farms |                    |                     | Percent control farms |                    |                     |
|--|--------------------|--------------------|---------------------|-----------------------|--------------------|---------------------|
|  |                    |                    |                     |                       |                    |                     |
| Small birds (finches, sparrows, starlings)   | 0                  | 0                  | 100                 | 0                     | 14                 | 86                  |
| Other  | 0                  | 0                  | 100                 | 0                     | 0                  | 100                 |
| Dead birds:  |                    |                    |                     |                       |                    |                     |
| Inside the barn  |                    | 0                  |                     |                       | 0                  |                     |
| Outside the barn   |                    | 0                  |                     |                       | 7                  |                     |
| Number of employees:   |                    |                    |                     |                       |                    |                     |
| 1  |                    | 38                 |                     |                       | 7                  |                     |
| 2  |                    | 25                 |                     |                       | 29                 |                     |
| 3 or more  |                    | 38                 |                     |                       | 64                 |                     |
| Frequency measures were required for workers entering the barns during the 14-day risk period: | <b>Always</b>      | <b>Some-times</b>  | <b>Never</b>        | <b>Always</b>         | <b>Some-times</b>  | <b>Never</b>        |
| Establish clean/dirty line   | 100                | 0                  | 0                   | 93                    | 0                  | 7                   |
| Shower   | 13                 | 0                  | 87                  | 0                     | 3                  | 97                  |
| Wash hands/hand sanitizer  | 37                 | 13                 | 50                  | 27                    | 17                 | 57                  |
| Different personnel for different barns  | 0                  | 0                  | 100                 | 13                    | 10                 | 77                  |
| Disposable coveralls   | 25                 | 0                  | 75                  | 23                    | 0                  | 77                  |
| Change clothing  | 62                 | 0                  | 38                  | 13                    | 13                 | 73                  |
| Change shoes/shoe covers   | 100                | 0                  | 0                   | 97                    | 3                  | 0                   |
| Liquid foot bath   | 100                | 0                  | 0                   | 83                    | 0                  | 17                  |
| Dry foot bath  | 0                  | 0                  | 100                 | 18                    | 0                  | 82                  |
| Scrub footwear   | 37                 | 0                  | 63                  | 37                    | 27                 | 37                  |
| Workers on the farm:   |                    |                    |                     |                       |                    |                     |
| Work on other company farms  |                    | 50                 |                     |                       | 50                 |                     |
| Employed by other poultry operations   |                    | 13                 |                     |                       | 17                 |                     |
| Own their own poultry  |                    | 0                  |                     |                       | 7                  |                     |
| Required to stay off farm at least 24 h after exposure to other poultry                        |                    | 13                 |                     |                       | 0                  |                     |
| Visitor log used   |                    | 71                 |                     |                       | 41                 |                     |
| Visitor type during the 14-day risk period:  | <b>Visitors</b>    | <b>Avg # times</b> | <b>Entered barn</b> | <b>Visitors</b>       | <b>Avg # times</b> | <b>Entered barn</b> |
| Federal/State veterinary or animal health worker   | 0                  |                    |                     | 0                     |                    |                     |
| Extension agent or university veterinarian   | 13                 |                    | 0                   | 0                     |                    |                     |
| Private or company veterinarian  | 13                 | 2                  | 25                  | 0                     |                    |                     |
| Company service person   | 88                 | 2                  | 88                  | 72                    | 2                  | 57                  |
| Nutritionist or feed company consultant  | 0                  |                    |                     | 0                     |                    |                     |
| Bird delivery personnel  | 0                  |                    |                     | 10                    | 1                  | 0                   |
| Vaccination crew   | 0                  |                    |                     | 0                     |                    |                     |
| Catch crew (bird removal)  | 25                 | 1                  | 14                  | 7                     | 1                  | 10                  |
| Feed delivery personnel  | 88                 | 8                  | 13                  | 97                    | 4                  | 8                   |
| Egg truck personnel (for breeder farms)  | 0                  |                    |                     | 0                     |                    |                     |

| Characteristic  | Percent case farms           |                                    |           | Percent control farms        |                                    |           |
|---|------------------------------|------------------------------------|-----------|------------------------------|------------------------------------|-----------|
|   |                              |                                    |           |                              |                                    |           |
| Litter delivery services  | 13                           | 1                                  | 0         | 23                           | 2                                  | 3         |
| Litter removal services (e.g., litter broker, litter disposal)                                    | 0                            |                                    |           | 0                            |                                    |           |
| Customer (private individual)   | 0                            |                                    |           | 0                            |                                    |           |
| Wholesaler, buyer, or dealer  | 0                            |                                    |           | 0                            |                                    |           |
| Renderer  | 0                            |                                    |           | 0                            |                                    |           |
| Occasional worker (e.g., family member, part-time help over holiday)                              | 50                           | 8                                  | 38        | 23                           | 2                                  | 20        |
| Construction workers, repair or maintenance person  | 0                            |                                    |           | 3                            | 1                                  | 0         |
| Other business visitors (including other producers, meter readers, package delivery (UPS), etc.)  | 25                           | 1                                  | 13        | 20                           | 1                                  | 0         |
| Other nonbusiness visitors (including neighbors, friends, family members, and school field trips) | 14                           | 14                                 | 0         | 7                            | 3                                  | 0         |
| Any visitors enter the barn   | 100                          |                                    |           | 57                           |                                    |           |
| Requirements for visitors who entered the barn during the 14-day risk period:                     | <b>Yes; verified at farm</b> | <b>Yes; visitor responsibility</b> | <b>No</b> | <b>Yes; verified at farm</b> | <b>Yes; visitor responsibility</b> | <b>No</b> |
| Change outer clothing   | 75                           | 25                                 | 0         | 25                           | 46                                 | 29        |
| Foot covers or change footwear  | 75                           | 25                                 | 0         | 37                           | 42                                 | 21        |
| Mask  | 37                           | 0                                  | 63        | 13                           | 4                                  | 83        |
| Hand sanitizer or gloves  | 75                           | 25                                 | 0         | 30                           | 39                                 | 30        |
| Not visit multiple farms in the same day  | 0                            | 0                                  | 100       | 4                            | 8                                  | 88        |
| Other   | 0                            | 0                                  | 100       | 0                            | 4                                  | 96        |
| Shared the following vehicles with another farm during the 14-day defined risk period:            |                              |                                    |           |                              |                                    |           |
| Company trucks/trailers (e.g., pickup truck, trailer with supplies, supervisor truck)             | 100                          |                                    |           | 86                           |                                    |           |
| Feed trucks   | 100                          |                                    |           | 90                           |                                    |           |
| Bird delivery (i.e., placing birds)   | 0                            |                                    |           | 20                           |                                    |           |
| Bird removal  | 11                           |                                    |           | 17                           |                                    |           |
| Egg removal (for breeder farms)   | 0                            |                                    |           | 0                            |                                    |           |
| Manure/litter hauling   | 0                            |                                    |           | 10                           |                                    |           |
| ATV/4-wheeler   | 22                           |                                    |           | 7                            |                                    |           |
| Other   | 0                            |                                    |           | 10                           |                                    |           |
| Equipment   | 11                           |                                    |           | 40                           |                                    |           |
| Litter heat treated   | 33                           |                                    |           | 27                           |                                    |           |
| Litter storage:   | 78                           |                                    |           | 40                           |                                    |           |
| Outside   | 0                            |                                    |           | 10                           |                                    |           |
| Closed shed   | 33                           |                                    |           | 13                           |                                    |           |
| Open shed   | 44                           |                                    |           | 23                           |                                    |           |
| Stored less than 30 yards from barn   | 60                           |                                    |           | 73                           |                                    |           |
| Stored litter accessible to:  |                              |                                    |           |                              |                                    |           |
| Wild birds  | 57                           |                                    |           | 58                           |                                    |           |

| Characteristic  | Percent case farms | Percent control farms |
|---|--------------------|-----------------------|
| Wild animals  | 57                 | 58                    |
| Domestic animals  | 57                 | 58                    |
| Litter disposal method:   |                    |                       |
| Composted on-farm   | 33                 | 11                    |
| Stored on-farm  | 11                 | 11                    |
| Applied to land on farm   | 11                 | 15                    |
| Taken off-site  | 89                 | 76                    |
| Manure or used litter from other farms spread on this farm  | 0                  | 3                     |
| Normal daily mortality (%):   |                    |                       |
| Less than 0.2   | 100                | 89                    |
| 0.2 or more   | 0                  | 11                    |
| Dead bird (daily mortality) disposal method:  |                    |                       |
| Composting on farm  | 67                 | 90                    |
| Burial  | 0                  | 0                     |
| Incineration  | 11                 | 0                     |
| Rendering   | 0                  | 0                     |
| Landfill  | 0                  | 0                     |
| Other off-farm  | 22                 | 10                    |
| If compost or burial, cover with:   |                    |                       |
| Soil  | 0                  | 7                     |
| Manure  | 100                | 78                    |
| Sawdust   | 0                  | 7                     |
| Wild birds or wild mammals observed around the dead bird collection area during the 14-day risk period: |                    |                       |
| Wild birds  | 63                 | 61                    |
| Wild mammals  | 50                 | 50                    |
| Common (off-farm) collection point for dead bird disposal   | 33                 | 31                    |

**APPENDIX B. BARN-LEVEL QUESTIONS - UPDATED**

Percentage of barns by barn characteristics

| Characteristic   | Percent case barns | Percent control barns |
|--|--------------------|-----------------------|
| Type(s) of poultry present in this barn:   |                    |                       |
| Brooder  | 0                  | 26                    |
| Grower toms  | 100                | 74                    |
| Grower hens  | 0                  | 4                     |
| Breeders   | 0                  | 0                     |
| Other  | 0                  | 0                     |
| Birds in this barn have outside access   | 11                 | 0                     |
| Number of birds placed in this barn:   |                    |                       |
| Fewer than 4,000   | 0                  | 4                     |
| 4,000 to 9,999   | 100                | 64                    |
| 10,000 or more   | 0                  | 32                    |
| Length of time birds in barn (weeks):  |                    |                       |
| Less than 4  | 0                  | 26                    |
| 4 to less than 8   | 25                 | 33                    |
| 8 to less than 12  | 38                 | 15                    |
| 12 or more   | 38                 | 26                    |
| Age of birds (weeks):  |                    |                       |
| Less than 8  | 0                  | 44                    |
| 8 to less than 14  | 25                 | 15                    |
| 14 or more   | 75                 | 41                    |
| Different stages of production (e.g., brooders and growers) present in this barn at the same time  | 11                 | 0                     |
| Another health concern in this flock during the defined risk period  | 44                 | 11                    |
| Flock being treated for a condition or health concern during the defined risk period   | 44                 | 18                    |
| Age of barn structure (yrs.):  |                    |                       |
| Less than 5  | 0                  | 4                     |
| 5 to less than 10  | 11                 | 25                    |
| 10 or more   | 89                 | 71                    |
| For barns more than 5 years old, last remodeled within 5 years   | 78                 | 22                    |
| How well the barn structure has been maintained:   |                    |                       |
| 1. Well (e.g., walls, curtains, and mud boards do not have holes, no visible daylight, the barn is tight and well insulated)   | 67                 | 75                    |
| 2. Moderate (e.g., barn could have rust or small holes, mud boards may be damaged, curtains may be torn or not in good repair, curtains may not close all the way, insulation may not be in good repair, the poly may be hanging from the ceiling) | 33                 | 25                    |

| Characteristic  |                       | Percent case barns        |                      |                       | Percent control barns     |                      |               |
|---|-----------------------|---------------------------|----------------------|-----------------------|---------------------------|----------------------|---------------|
| 3. Poor (e.g., holes in walls and mud boards are apparent, tin is rusted, may have leaks in roof, there might be some holes large enough for wild birds to enter, multiple areas with daylight visible, insulation may be hanging from the ceiling) |                       | 0                         |                      |                       | 0                         |                      |               |
| Type of ventilation used for this barn during the 14-day defined risk period  |                       |                           |                      |                       |                           |                      |               |
| Curtain ventilated  |                       | 33                        |                      |                       | 32                        |                      |               |
| Environmental control/tunnel ventilation  |                       | 56                        |                      |                       | 54                        |                      |               |
| Side doors (i.e., tip outs)   |                       | 11                        |                      |                       | 11                        |                      |               |
| Other   |                       | 0                         |                      |                       | 4                         |                      |               |
| Intake air filtered   |                       | 0                         |                      |                       | 0                         |                      |               |
| Needed to repair or replace any feed tank lids in the past 14 days  |                       | 0                         |                      |                       | 0                         |                      |               |
| Any feed tank lids open in the past 14 days   |                       | 0                         |                      |                       | 0                         |                      |               |
| Ground surface immediately surrounding (within 1 yard) this barn (excluding vehicle approach and loading area):   |                       |                           |                      |                       |                           |                      |               |
| Gravel or hard surface  |                       | 56                        |                      |                       | 67                        |                      |               |
| Dirt  |                       | 0                         |                      |                       | 7                         |                      |               |
| Short grass   |                       | 44                        |                      |                       | 26                        |                      |               |
| Tall grass or brush   |                       | 0                         |                      |                       | 0                         |                      |               |
| Hard-surface entry pad (e.g., concrete, asphalt):   |                       |                           |                      |                       |                           |                      |               |
| Cleaned and disinfected   |                       | 33                        |                      |                       | 68                        |                      |               |
| Cleaned only  |                       | 11                        |                      |                       | 14                        |                      |               |
| Not cleaned or disinfected  |                       | 33                        |                      |                       | 7                         |                      |               |
| No hard-surface entry pad   |                       | 22                        |                      |                       | 11                        |                      |               |
| Frequency the following were used in this barn during the 14-day defined risk period:   | <b>Used regularly</b> | <b>Not used regularly</b> | <b>Not available</b> | <b>Used regularly</b> | <b>Not used regularly</b> | <b>Not available</b> | <b>pvalue</b> |
| Locks on the doors  | 11                    | 44                        | 44                   | 4                     | 74                        | 22                   | .25           |
| A service room that personnel must enter through that separates "outside area" from "inside area"   | 100                   | 0                         | 0                    | 86                    | 0                         | 14                   | .55           |
| Changing area for employees   | 89                    | 0                         | 11                   | 68                    | 11                        | 21                   | .41           |
| A shower for employees  | 11                    | 0                         | 89                   | 0                     | 7                         | 93                   |               |
| Cool cell pads  | 0                     | 22                        | 78                   | 11                    | 25                        | 64                   | .55           |
| Misters   | 0                     | 89                        | 11                   | 0                     | 48                        | 52                   | .03           |
| Type of footbath in use at this barn:   |                       |                           |                      |                       |                           |                      |               |
| Dry (i.e., powdered or particulate)   |                       | 0                         |                      |                       | 18                        |                      |               |
| Liquid  |                       | 100                       |                      |                       | 82                        |                      |               |
| Other   |                       | 0                         |                      |                       | 0                         |                      |               |
| None  |                       | 0                         |                      |                       | 0                         |                      |               |
| Dry footbath changed at least 1x/week   |                       | -                         |                      |                       | 60                        |                      |               |

| Characteristic   | Percent case barns | Percent control barns |
|--|--------------------|-----------------------|
| Liquid footbath changed at least 1x/day  | 0                  | 0                     |
| Type of litter used in this barn:  |                    |                       |
| Wood shavings  | 89                 | 75                    |
| Hulls (e.g., oat, rice, sunflower, other)  | 11                 | 25                    |
| Other  | 0                  | 0                     |
| Litter is:   |                    |                       |
| Bagged (i.e., baled)   | 0                  | 4                     |
| Bulk (i.e., load from shavings mill)   | 100                | 96                    |
| Litter was "tilled" after it was placed in the barn  | 67                 | 75                    |
| Litter was added to the barn during the defined risk period                                      | 0                  | 12                    |
| Partial clean-out performed in this barn (during the 14-day defined risk period)                 | 0                  | 4                     |
| Last full clean-out:   |                    |                       |
| Prior to this flock  | 67                 | 68                    |
| Two flocks ago   | 22                 | 21                    |
| Three or more flocks ago   | 11                 | 11                    |
| If a full cleanout was performed, person who performed the full cleanout:                        |                    |                       |
| Grower   | 78                 | 79                    |
| Contractor   | 22                 | 21                    |
| Partial load-out occurred while this flock was present   | 0                  | 7                     |
| Wild birds seen in this barn during the 14-day defined risk period                               | 0                  | 0                     |
| Types of people who entered this barn during the 14-day defined risk period:                     |                    |                       |
| Federal/State veterinary or animal health worker   | 0                  | 0                     |
| Extension agent or university veterinarian   | 0                  | 0                     |
| Private or company veterinarian  | 29                 | 4                     |
| Company service person   | 88                 | 61                    |
| Nutritionist or feed company consultant  | 0                  | 0                     |
| Bird delivery personnel  | 0                  | 0                     |
| Vaccination crew   | 0                  | 0                     |
| Catch crew (bird removal)  | 0                  | 0                     |
| Feed delivery personnel  | 0                  | 7                     |
| Egg truck personnel (for breeder farms)  | 0                  | 0                     |
| Litter delivery services   | 0                  | 0                     |
| Litter removal services (e.g., litter broker, litter disposal)                                   | 0                  | 0                     |
| Customer (private individual)  | 0                  | 0                     |
| Wholesaler, buyer, or dealer   | 0                  | 0                     |
| Renderer   | 0                  | 0                     |
| Occasional worker (e.g., family member, part time help over holiday)                             | 38                 | 21                    |
| Construction workers, repair or maintenance person   | 0                  | 0                     |
| Other business visitors (including other producers, meter readers, package delivery (UPS), etc.) | 13                 | 0                     |

| Characteristic   | Percent case barns | Percent control barns |
|--|--------------------|-----------------------|
| Other nonbusiness visitors (including neighbors, friends, family members, and school field trips)  | 0                  | 0                     |
| Any visitor enter the barn   | 100                | 68                    |
| If controller information is available median percentage of time that curtains were open and median numbers of days open or partially open during the defined risk period: |                    |                       |
| % time   | 0                  | 0                     |
| # days   | 0                  | 0                     |
| Median minimum and maximum temperatures in the barn during the 14-day defined risk period?   |                    |                       |
| Minimum  | 55                 | 61                    |
| Maximum  | 65                 | 69                    |
| Median distance this barn (in yards) from:   |                    |                       |
| Dead bird disposal/holding area including carcass bin for rendering  | 59                 | 100                   |
| Litter compost (for barns where litter is composted)   | 50                 | 100                   |
| Nearest road   | 100                | 100                   |

## **APPENDIX C. HPAI CASE CONTROL QUESTIONNAIRE**

HPAI turkeys, version 4, January 15, 2016



# HPAI Case Control Questionnaire

## Turkeys

Animal and Plant Health Inspection Service

Veterinary Service:

National Animal Health Monitoring System

2150 Centre Ave., Bldg B  
Fort Collins, CO 80526

Form Approved  
OMB Number 0579-0376  
Approval Expires:  
9/30/2017

Study ID: \_\_\_\_\_ frmid

EMRS (premises) ID: \_\_\_\_\_ premid

Date (mm/dd/yy): \_\_\_\_\_ date

**Interviewer instruction:** Bring satellite image of farm to interview. Enter Lat and Long at farm gate.

### A. PREMISES INFORMATION

Farm name: \_\_\_\_\_ frmname

Farm address: \_\_\_\_\_ frmadd

County: \_\_\_\_\_ frmcty      Lat \_\_\_\_\_ lat      Long \_\_\_\_\_ long

1. Company flock supervisor name: \_\_\_\_\_ t101

Phone: \_\_\_\_\_ t102      Cell phone: \_\_\_\_\_ t103      Email: \_\_\_\_\_ t104

2. Farm manager contact name: \_\_\_\_\_ t105

Phone: \_\_\_\_\_ t106      Cell phone: \_\_\_\_\_ t107      Email: \_\_\_\_\_ t108

3. Flock veterinarian: \_\_\_\_\_ t109

Phone: \_\_\_\_\_ t110      Cell phone: \_\_\_\_\_ t111      Email: \_\_\_\_\_ t112

### B. INTERVIEWER INFORMATION

Interviewer name/organization: \_\_\_\_\_ intrname

Interviewee name/organization: \_\_\_\_\_ intename



Animal and Plant Health Inspection Service

Veterinary Services

# HPAI Case Control Questionnaire

## Turkeys

National Animal Health Monitoring System

2150 Centre Ave., Bldg B  
Fort Collins, CO 80526

Form Approved  
OMB Number 0579-0376  
Approval Expires:  
9/30/2017

Study ID: \_\_\_\_\_ frmid

Date (mm/dd/yy): \_\_\_\_\_ date

### INSTRUCTIONS

State and local poultry organizations [fill in cooperators here] and the U.S. Department of Agriculture APHIS (USDA APHIS) are conducting a case-control study as part of the highly pathogenic avian influenza (HPAI) investigation efforts to identify factors that may contribute to transmission of H5N2 influenza virus to poultry.

We are asking you to fill out this survey, which includes questions about things done daily on the farm, facility and premises condition, deliveries to the farm, and ill birds. We ask about a 2-week (14-day) period on the farm starting on a particular date that we will provide. It might be difficult to remember back that far, so please use a pocket calendar or other agenda manager and any feed and other delivery records that might be available to you.

| Term     | Case definition  | Control definition   |
|----------|--|--|
| Premises | Farm location with flocks confirmed to be HPAI H5N2 infected by NVSL, including all barns and buildings, even if not all barns and buildings contain infected birds. | Turkey farm location with no infected birds in any barn or building, in close proximity (less than 10 miles) to the case farm. (If case farm is a turkey breeder, select a noninfected turkey breeder as the control.) |
| Barn     | Barn or building that houses HPAI H5N2-infected birds.   | On control premises, a barn or building that does not house any infected birds.  |

### Dates of study focus:

**Case farms** answer questions for the timeframe of 14 days prior to the onset of clinical signs or increased mortality. All questions that ask about the “defined risk period” refer to this time period.

**Control farms** answer questions for the timeframe of 14 days prior to date of first detection on the matched case farm. All questions that ask about the “defined risk period” are referring to this time period.

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0579-0376. The time required to complete this information collection is estimated to average 1 hour per response, including the time to review instructions, search existing data resources, gather the data needed, and complete and review the information collected.

**NAHMS-348**  
**SEP 2015**

### A. CASE OR CONTROL

1. Is this a case or control farm? t201

- <sub>1</sub> Case [Go to question 2.]
- <sub>3</sub> Control [Go to question 3.]

2. If this is a **case** farm,

- a. When were clinical signs or increased mortality first observed?  
This is what we will refer to as the **reference date** ..... t202 \_\_\_\_\_ mm/dd/yy
  - b. Enter the date 14 days prior to the date of first detection  
(clarifying timeframe of study focus) .....t203 \_\_\_\_\_ mm/dd/yy
- All questions regarding the “defined risk period” refer to the 14 days prior to this reference date (i.e., the time between “a” and “b”).**
- c. How many birds were on this farm on this reference date?.....t204 \_\_\_\_\_ # birds
  - d. On the reference date, was this farm in an existing control zone? .... t205..... <sub>1</sub> Yes <sub>3</sub> No
  - e. When was the flock diagnosed as positive by laboratory confirmation? .....t206 \_\_\_\_\_ mm/dd/yy
  - f. As of today, how many of the barns on this farm have been confirmed or are suspected of being infected with HPAI?.....t207 \_\_\_\_\_ # barns

[Go to section B.]

3. If this is a **control** farm,

- a. Enter reference date  
(enter date of matched case farm prior to interview) .....t208 \_\_\_\_\_ mm/dd/yy
  - b. Enter the date 14 days prior to the reference date .....t209 \_\_\_\_\_ mm/dd/yy
- All questions regarding the “defined risk period” refer to the 14 days prior to this reference date (i.e., the time between “a” and “b”).**
- c. How many birds were on this farm on this reference date?.....t210 \_\_\_\_\_ # birds
  - d. Is this farm located in a control zone today? .....t211 <sub>1</sub> Yes <sub>3</sub> No
- If Yes, how long has it been in a control zone? ..... t212d/t212w \_\_\_\_\_ days OR \_\_\_\_\_ weeks

**B. PREMISES DESCRIPTION**

1. Is this a: *[Check one only.]* t301
- <sub>1</sub> Company farm?
  - <sub>2</sub> Contract farm?
  - <sub>3</sub> Lease farm?
  - <sub>4</sub> Independent farm?
2. What stage(s) of turkey production is on this farm?
- a. Brooder ..... t302 <sub>1</sub> Yes <sub>3</sub> No
  - b. Grower ..... t303 <sub>1</sub> Yes <sub>3</sub> No
  - c. Breeder ..... t304 <sub>1</sub> Yes <sub>3</sub> No
  - d. Other (specify: \_\_\_\_\_) t305oth ..... t305 <sub>1</sub> Yes <sub>3</sub> No

**[If question 2a OR question 2b = No, SKIP to question 4.]**

3. Are brooders and growers: *[Check one only.]* t306
- <sub>1</sub> In the barn at the same time?
  - <sub>2</sub> In the same barn but at different times (e.g., placed as brooders and remain through grow-out)?
  - <sub>3</sub> In different barns?
4. What is the sex of the market type on this farm? *[Check one only.]* t307
- <sub>1</sub> Hens (HH or LH)
  - <sub>2</sub> Toms (FP)
  - <sub>3</sub> Both hens and toms (i.e., multiple market classes)
  - <sub>4</sub> Breeder hens
  - <sub>5</sub> Breeder toms
5. Is this farm multiple age or single age? *[Check one only.]* t308
- <sub>1</sub> Multiple age
  - <sub>2</sub> Single age
6. What other type(s) of poultry is present on this farm?
- a. Broiler..... t309 <sub>1</sub> Yes <sub>3</sub> No
  - b. Layer..... t310 <sub>1</sub> Yes <sub>3</sub> No
  - c. Other (specify: \_\_\_\_\_) t311oth ..... t311 <sub>1</sub> Yes <sub>3</sub> No

7. Is this farm certified organic? .....t312 <sub>1</sub> Yes <sub>3</sub> No
8. Is this facility enrolled in NPIP? .....t313 <sub>1</sub> Yes <sub>3</sub> No
9. How many barns are on this farm? .....t314 \_\_\_\_\_ # barns
10. Do any birds on the farm have access to the outdoors?.....t315 <sub>1</sub> Yes <sub>3</sub> No
11. Are the following water body type(s) visible or within 350 yards (about three football fields) of this farm?
- a. Pond .....t316 <sub>1</sub> Yes <sub>3</sub> No
- b. Lake .....t317 <sub>1</sub> Yes <sub>3</sub> No
- c. Stream .....t318 <sub>1</sub> Yes <sub>3</sub> No
- d. River .....t319 <sub>1</sub> Yes <sub>3</sub> No
- e. Wetland or swamp .....t320 <sub>1</sub> Yes <sub>3</sub> No
- f. Wastewater lagoon .....t321 <sub>1</sub> Yes <sub>3</sub> No
- g. Other (specify: \_\_\_\_\_) t322oth .....t322 <sub>1</sub> Yes <sub>3</sub> No
12. For those water bodies within 350 yards, approximately how many of the following types of waterfowl were seen on the water during the 14-day defined risk period?
- a. Ducks ..... t323 <sub>1</sub> None <sub>2</sub> Tens <sub>3</sub> Hundreds <sub>4</sub> Thousands <sub>5</sub> Don't know
- b. Geese..... t324 <sub>1</sub> None <sub>2</sub> Tens <sub>3</sub> Hundreds <sub>4</sub> Thousands <sub>5</sub> Don't know
- c. Shorebirds  
(e.g., wading birds, gulls)..... t325 <sub>1</sub> None <sub>2</sub> Tens <sub>3</sub> Hundreds <sub>4</sub> Thousands <sub>5</sub> Don't know
- d. Other  
(specify: \_\_\_\_\_) t326oth ..... t326 <sub>1</sub> None <sub>2</sub> Tens <sub>3</sub> Hundreds <sub>4</sub> Thousands <sub>5</sub> Don't know
13. What is the approximate distance (in yards) to the closest field where crops or hay are harvested? ..... t327 \_\_\_\_\_ yards
14. What crop was last grown in this field? [Check one only.] t328
- <sub>1</sub> Corn
- <sub>2</sub> Soybeans
- <sub>3</sub> Alfalfa or grass intended for livestock feed
- <sub>4</sub> Other (specify: \_\_\_\_\_) t328oth
15. Was this field tilled last fall? ..... t329 <sub>1</sub> Yes <sub>3</sub> No <sub>4</sub> Don't know

16. Was this field actively worked (e.g., tilled, disked, hay harvested, trees cut, row crops harvested) during the 14-day defined risk period? ..... t330 <sub>1</sub> Yes <sub>3</sub> No <sub>4</sub> Don't know
17. For this closest field, approximately how many of the following types of waterfowl were seen during the 14-day defined risk period?
- a. Ducks ..... t331 <sub>1</sub> None <sub>2</sub> Tens <sub>3</sub> Hundreds <sub>4</sub> Thousands <sub>5</sub> Don't know
  - b. Geese..... t332 <sub>1</sub> None <sub>2</sub> Tens <sub>3</sub> Hundreds <sub>4</sub> Thousands <sub>5</sub> Don't know
  - c. Shorebirds (e.g., wading birds, gulls)..... t333 <sub>1</sub> None <sub>2</sub> Tens <sub>3</sub> Hundreds <sub>4</sub> Thousands <sub>5</sub> Don't know
  - d. Other (specify: \_\_\_\_\_) t334oth ..... t334 <sub>1</sub> None <sub>2</sub> Tens <sub>3</sub> Hundreds <sub>4</sub> Thousands <sub>5</sub> Don't know
18. What other types of animals are present on the farm premises?
- a. Beef cattle .....t335 <sub>1</sub> Yes <sub>3</sub> No
  - b. Dairy cattle .....t336 <sub>1</sub> Yes <sub>3</sub> No
  - c. Horses.....t337 <sub>1</sub> Yes <sub>3</sub> No
  - d. Sheep.....t338 <sub>1</sub> Yes <sub>3</sub> No
  - e. Goats .....t339 <sub>1</sub> Yes <sub>3</sub> No
  - f. Pigs .....t340 <sub>1</sub> Yes <sub>3</sub> No
  - g. Dogs .....t341 <sub>1</sub> Yes <sub>3</sub> No
  - h. Cats .....t342 <sub>1</sub> Yes <sub>3</sub> No
  - i. Poultry or domesticated waterfowl (noncommercial) .....t343 <sub>1</sub> Yes <sub>3</sub> No
  - j. Other (specify: \_\_\_\_\_) t344oth.....t344 <sub>1</sub> Yes <sub>3</sub> No
19. What is the water source for poultry?
- a. Municipal.....t345 <sub>1</sub> Yes <sub>3</sub> No
  - b. Well .....t346 <sub>1</sub> Yes <sub>3</sub> No
  - c. Surface water (e.g., pond).....t347 <sub>1</sub> Yes <sub>3</sub> No
  - d. Other (specify: \_\_\_\_\_) t348oth.....t348 <sub>1</sub> Yes <sub>3</sub> No
20. Are the following water treatments used in the drinking water for the poultry on this farm?
- a. Chlorination.....t349 <sub>1</sub> Yes <sub>3</sub> No
  - b. Acidifiers.....t350 <sub>1</sub> Yes <sub>3</sub> No
  - c. Iodine.....t351 <sub>1</sub> Yes <sub>3</sub> No
  - d. Peroxide .....t352 <sub>1</sub> Yes <sub>3</sub> No
  - e. Other (specify: \_\_\_\_\_) t353oth .....t353 <sub>1</sub> Yes <sub>3</sub> No

21. Are windbreaks present on this farm?  
 If Yes, what is the distance (in yards) from the windbreak to the closest poultry barn?

| Windbreak type                            | Present?   | If Yes, distance to closest poultry barn |           |
|---|--|--|-----------|
| a. Evergreen or juniper                   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ yards                              | t354/t357 |
| b. Deciduous tree                         | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ yards                              | t355/t358 |
| c. Structural (e.g., hill, natural break) | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ yards                              | t356/t359 |

22. Excluding driveways on farm, what is the distance (in yards or miles) from this farm to the nearest public gravel or dirt road? .....t360y/t360m \_\_\_\_\_yards OR \_\_\_\_\_ miles
23. What is the direction from this public roadway to the farm?.....t361 \_\_\_\_\_ direction

**C. FARM BIOSECURITY**

1. Is there a house with people living in it on the property? .....t401 <sub>1</sub> Yes <sub>3</sub> No

**[If question 1 = No, SKIP to question 3.]**

2. Is there a common drive entrance to farm and residence? .....t402 <sub>1</sub> Yes <sub>3</sub> No

3. How many entrances are there to the farm that could provide access to the poultry area? .....t403 \_\_\_\_\_ #

4. What best describes the road surface on this farm that vehicles coming onto the operation drive on? *[Check one only.]* .....t404

- <sub>1</sub> Hard top/asphalt
- <sub>2</sub> Gravel
- <sub>3</sub> Dirt
- <sub>4</sub> Other (specify: \_\_\_\_\_) t404oth

5. In general, do the following types of vehicles:

| Codes for question 5                       |
|--|
| 1 = come to the perimeter of the farm only |
| 2 = enter the farm but not near the barns  |
| 3 = come near the barns                    |
| 4 = do not come at all                     |

**Enter the codes that apply.**

- a. Garbage/dumpster pick up?.....t405      \_\_\_\_\_ code
  - b. Propane delivery?.....t406      \_\_\_\_\_ code
  - c. Feed delivery? .....t407      \_\_\_\_\_ code
  - d. Renderer?.....t408      \_\_\_\_\_ code
  - e. Company personnel (e.g., catch/vaccination crew, barn workers, service person, veterinarian)?.....t409      \_\_\_\_\_ code
  - f. Other business visitors (e.g., meter reader, repairman)? .....t411      \_\_\_\_\_ code
6. Is there a gate to this farm entrance? .....t412      <sub>1</sub> Yes <sub>3</sub> No

**[If question 6 = No, SKIP to question 8.]**

- 7. Is the gate secured/locked? .....t413      <sub>1</sub> Yes <sub>3</sub> No
- 8. Is the farm area perimeter surrounded by a security fence?.....t414      <sub>1</sub> Yes <sub>3</sub> No
- 9. How frequently is vegetation mowed/bush hogged on the premises (answer for when vegetation is present, e.g., spring and summer).....t415      \_\_\_\_\_ times/month
- 10. Was there a wash station/spray area being used for vehicles during the 14-day defined risk period? .....t416      <sub>1</sub> Yes <sub>3</sub> No

**[If question 10 = No, SKIP to question 12.]**

11. For wash station/spray area:

- a. Is it located on the farm? .....t417 <sub>1</sub> Yes <sub>3</sub> No
- b. Are the tires washed?.....t418 <sub>1</sub> Yes <sub>3</sub> No
- c. Is the vehicle exterior washed?.....t419 <sub>1</sub> Yes <sub>3</sub> No
- d. Is the vehicle interior cleaned (e.g., floor mats).....t420 <sub>1</sub> Yes <sub>3</sub> No
- e. Which vehicles are washed:
  - i. Worker vehicles?.....t421 <sub>1</sub> Yes <sub>3</sub> No
  - ii. Feed trucks? .....t422 <sub>1</sub> Yes <sub>3</sub> No
  - iii. Vehicles delivering/removing birds?.....t423 <sub>1</sub> Yes <sub>3</sub> No
  - iv. Other? (specify: \_\_\_\_\_) t424oth .....t424 <sub>1</sub> Yes <sub>3</sub> No
- f. What disinfectant is used? \_\_\_\_\_ t425
- g. Was the wash station: [Check one only.] t426
  - <sub>1</sub> Recently put into use as a response to heightened biosecurity concerns?
  - <sub>2</sub> A permanent station (i.e., in use prior to the HPAI incident)?

12. Did workers and visitors always, sometimes, or never park in a restricted area away from the poultry barns during the 14-day defined risk period?

- a. Workers .....t427 <sub>1</sub> Always <sub>2</sub> Sometimes <sub>3</sub> Never
- b. Visitors.....t428 <sub>1</sub> Always <sub>2</sub> Sometimes <sub>3</sub> Never

13. What pest and wildlife control measures were used on this farm during the 14-day defined risk period?

- a. Rat and mouse bait stations? .....t429 <sub>1</sub> Yes <sub>3</sub> No  
 If Yes, how frequently are they checked? .....t430 \_\_\_\_\_ times/month
- b. Beetle control? .....t431 <sub>1</sub> Yes <sub>3</sub> No  
 If Yes, what type was used?
  - i. Sprays.....t432 <sub>1</sub> Yes <sub>3</sub> No
  - ii. Boric acid.....t433 <sub>1</sub> Yes <sub>3</sub> No
  - iii. Baits.....t434 <sub>1</sub> Yes <sub>3</sub> No
  - iv. Other (specify: \_\_\_\_\_) t435oth.....t435 <sub>1</sub> Yes <sub>3</sub> No

c. Fly control (other than manure removal)? ..... b436 <sub>1</sub> Yes <sub>3</sub> No

If Yes, what type was used?

- i. Residual spray .....t437 <sub>1</sub> Yes <sub>3</sub> No
- ii. Baits.....t438 <sub>1</sub> Yes <sub>3</sub> No
- iii. Larvacide (spot treatment) .....t439 <sub>1</sub> Yes <sub>3</sub> No
- iv. Larvacide in feed .....t440 <sub>1</sub> Yes <sub>3</sub> No
- v. Space sprays/fogger .....t441 <sub>1</sub> Yes <sub>3</sub> No
- vi. Biological predators .....t442 <sub>1</sub> Yes <sub>3</sub> No
- vii. Other (specify: \_\_\_\_\_) t443oth.....t443 <sub>1</sub> Yes <sub>3</sub> No

14. How often were rodents observed in the poultry barns during the 14-day defined risk period?

[Check one only.]

t444

- <sub>1</sub> Frequently (e.g., daily)
- <sub>2</sub> Occasionally (e.g., weekly)
- <sub>3</sub> Never

15. What was the intensity of beetles observed in the poultry barns during the 14-day defined risk period?

t445

[Check one only.]

- <sub>1</sub> High
- <sub>2</sub> Medium
- <sub>3</sub> Low
- <sub>4</sub> Never

16. What was the intensity of flies observed in the poultry barns during the 14-day defined risk period?

t446

[Check one only.]

- <sub>1</sub> High
- <sub>2</sub> Medium
- <sub>3</sub> Low

17. Were wild mammals, such as raccoons, opossums, coyotes, or foxes (or evidence of their presence), seen in or around poultry barns during the 14-day defined risk period? .....

t447

<sub>1</sub> Yes <sub>3</sub> No

18. During the 14-day defined risk period, prior to feeding, how frequently did wild birds, wild animals, and rodents have access to poultry feed (i.e., feed spillage, open bag, cover left open)?

|   | Always/<br>nearly always              | Most of<br>the time                   | Sometimes                             | Never                                 |      |
|---|---------------------------------------|---------------------------------------|---------------------------------------|---------------------------------------|------|
| a. Wild birds   | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t448 |
| b. Wild animals such as raccoons, opossums, coyotes, or foxes | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t449 |
| c. Rodents  | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t450 |

19. Describe the protocol or plan for when feed spills on your farm: t451

\_\_\_\_\_

20. What form of feed is fed to the poultry?

- a. Mash.....t452 <sub>1</sub> Yes <sub>3</sub> No
- b. Pellets.....t453 <sub>1</sub> Yes <sub>3</sub> No
- c. Other (specify: \_\_\_\_\_) t454oth.....t454 <sub>1</sub> Yes <sub>3</sub> No

21. Is the feed treated with formaldehyde (i.e., Termin-8)?.....t455 <sub>1</sub> Yes <sub>3</sub> No

22. Is the feed heat treated?.....t456 <sub>1</sub> Yes <sub>3</sub> No

**D. WILD BIRDS**

1. How frequently were the following types of wild birds seen on the farm, but outside of the barns (within 100 yards), during the 14-day defined risk period?

| Bird type   | Often                                 | Sometimes                             | Never                                 |      |
|---|---------------------------------------|---------------------------------------|---------------------------------------|------|
| a. Waterfowl (e.g., ducks, geese)                             | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t501 |
| b. Gulls  | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t502 |
| c. Small perching birds (e.g., sparrows, starlings, swallows) | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t503 |
| d. Blackbirds and crows                                       | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t504 |
| e. Other water birds (e.g., egrets, cormorants)               | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t505 |
| f. Wild turkeys, pheasants, quail                             | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t506 |
| g. Raptors (e.g., eagles, hawks, owls)                        | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t507 |
| h. Pigeons and doves  | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t508 |
| i. Other (specify: _____) t509oth                             | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t509 |

2. How frequently were the following types of wild birds seen in the barns during the 14-day defined risk period?

| Bird type   | Often                                 | Sometimes                             | Never                                 |      |
|---|---------------------------------------|---------------------------------------|---------------------------------------|------|
| a. Large birds (e.g., pigeons, crows)               | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t510 |
| b. Small birds (e.g., finches, sparrows, starlings) | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t511 |
| c. Other (specify: _____) t512oth                   | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t512 |

3. Did you observe any of the following types of **dead** wild birds in the barns or **outside** of the barns during the 14-day defined risk period?

| Dead bird type                                      | Inside the barns?  | Outside the barns?   |           |
|---|--|--|-----------|
| a. Large birds (e.g., pigeons, crows)               | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t513/t516 |
| b. Small birds (e.g., finches, sparrows, starlings) | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t514/t517 |
| c. Other (specify: _____) t515oth                   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t515/t518 |

### E. FARM HELP/WORKERS

Questions in this section refer to persons such as the producer, employees, farm help, crews, etc.

- What is the total number of employees working on this farm that have access to or directly work with poultry (including family, both paid and unpaid)?.....t601 \_\_\_\_\_ #
- During the 14-day defined risk period, were the following measures always/nearly always, most of the time, sometimes, or never required for workers entering the poultry barns?

| Measure  | Always/<br>nearly<br>always           | Most of the<br>time                   | Sometimes                             | Never                                 |      |
|--|---------------------------------------|---------------------------------------|---------------------------------------|---------------------------------------|------|
| a. An established clean/dirty line   | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t602 |
| b. Shower  | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t603 |
| c. Wash hands or use hand sanitizer before entering and/or before leaving the barn | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t604 |
| d. Different personnel for different barns   | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t605 |
| e. Wear disposable coveralls   | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t606 |
| f. Change of clothing (washable)   | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t607 |
| g. Change of shoes or use of shoe covers   | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t608 |
| h. Foot bath (liquid)  | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t609 |
| i. Foot bath (dry)   | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t610 |
| j. Scrub footwear (bucket and brush)   | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | <input type="checkbox"/> <sub>4</sub> | t611 |

- 3. Do workers on this farm work on other company farms? .....t612 <sub>1</sub> Yes <sub>3</sub> No
- 4. Are workers or members of their household employed by other poultry operations, rendering plants, or processing plants? .....t613 <sub>1</sub> Yes <sub>3</sub> No
- 5. Do any employees own their own poultry, including small backyard flocks? ..... t614 <sub>1</sub> Yes <sub>3</sub> No <sub>4</sub> Don't know
- 6. Are employees required to stay off farm after exposure to other poultry? .....t615 <sub>1</sub> Yes <sub>3</sub> No  
If Yes, for how long (hours)?.....t616 \_\_\_\_\_ hours

**F. FARM VISITORS**

- 1. Is a visitor log used to record visitor traffic onto the farm?.....t701 <sub>1</sub> Yes <sub>3</sub> No
- 2. Did any of the following types of people visit the farm during the 14-day defined risk period? If Yes, how many times did they visit and did they enter the poultry barn?

| Visitor type  | Did they visit the farm?   | If Yes...                      |  |                |
|---|--|--------------------------------|--|----------------|
|   |  | How many times did they visit? | Did this visitor enter the poultry barn?   |                |
| a. Federal/State veterinary or animal health worker               | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t702/t721/t740 |
| b. Extension agent or university veterinarian                     | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t703/t722/t741 |
| c. Private or company veterinarian                                | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t704/t723/t742 |
| d. Company service person   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t705/t724/t743 |
| e. Nutritionist or feed company consultant                        | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t706/t725/t744 |
| f. Bird delivery personnel  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t707/t726/t745 |
| g. Vaccination crew   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t708/t727/t746 |
| h. Catch crew (bird removal)                                      | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t709/t728/t747 |
| i. Feed delivery personnel  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t710/t729/t748 |
| j. Egg truck personnel (for breeder farms)                        | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t711/t730/t749 |
| k. Litter delivery services                                       | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t712/t731/t750 |
| l. Litter removal services (e.g., litter broker, litter disposal) | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t713/t732/t751 |
| m. Customer (private individual)                                  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t714/t733/t752 |
| n. Wholesaler, buyer, or dealer                                   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t715/t734/t753 |
| o. Renderer   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t716/t735/t754 |

|  |  |                |  |                |
|--|--|----------------|--|----------------|
| p. Occasional worker (e.g., family member, part-time help over holiday)                              | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t717/t736/t755 |
| q. Construction workers, repair or maintenance personnel   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t718/t737/t756 |
| r. Other business visitors (including other producers, meter readers, package delivery (UPS), etc.)  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t719/t738/t757 |
| s. Other nonbusiness visitors (including neighbors, family members, friends, and school field trips) | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | _____ # visits | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t720/t739/t758 |

3. For those visitors who entered the poultry barn during the 14-day defined risk period, did you require the following?

|   | Yes, verified at farm                 | Yes, visitor responsibility           | No                                    |      |
|---|---------------------------------------|---------------------------------------|---------------------------------------|------|
| a. Change of outer clothing/ farm specific clothing | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t759 |
| b. Foot covers or change of footwear                | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t760 |
| c. Mask   | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t761 |
| d. Hand sanitizing or gloves                        | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t762 |
| e. Not visit multiple farms in the same day         | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t763 |
| f. Other (specify: _____) t764oth                   | <input type="checkbox"/> <sub>1</sub> | <input type="checkbox"/> <sub>2</sub> | <input type="checkbox"/> <sub>3</sub> | t764 |

### G. FARM VEHICLES AND EQUIPMENT

1. Were the following vehicles shared with another farm during the 14-day defined risk period?

| Vehicle type   | Shared with another farm?  |      |
|--|--|------|
| a. Company trucks/trailers (e.g., pickup truck, trailer with supplies, supervisor truck, etc.) | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t801 |
| b. Feed trucks   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t802 |
| c. Bird delivery vehicles (i.e., placing birds)  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t803 |
| d. Bird removal vehicles   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t804 |
| e. Egg removal vehicles (for breeder farms)  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t805 |
| f. Manure/litter hauling   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t806 |
| g. ATV/4-wheeler   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t807 |
| h. Other (specify: _____) t808oth  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t808 |

2. Were the following pieces of equipment shared with another farm during the 14-day defined risk period?

| Equipment type                       | Shared with another farm?  |      |
|--------------------------------------|--|------|
| a. Gates/panels                      | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t810 |
| b. Lawn mowers                       | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t811 |
| c. Live haul loaders                 | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t812 |
| d. Catch pens                        | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t813 |
| e. Scales for weighing birds         | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t814 |
| f. Vaccination equipment             | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t815 |
| g. Pressure sprayers/washers/foamers | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t816 |
| h. Skid-steer loaders                | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t817 |
| i. Litter/manure handling            | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t818 |
| j. Tillers/de-caking equipment       | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t819 |
| j. Other (specify: _____) t820oth    | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t820 |

**H. LITTER HANDLING**

1. What was the last day litter was brought onto the farm? .....t901 \_\_\_\_\_ mm/dd/yy
  
2. For the last litter (bedding) delivery, who brought the litter onto the farm?  
*[Check one only.]* t902
  - <sub>1</sub> Company personnel
  - <sub>2</sub> Litter provider
  - <sub>3</sub> Other? (specify: \_\_\_\_\_) t902oth
  
3. Is the litter heat treated prior to delivery? ..... t903 <sub>1</sub> Yes <sub>3</sub> No <sub>4</sub> Don't know
  
4. Is litter stored on the farm prior to use:
  - a. Outside? .....t904 <sub>1</sub> Yes <sub>3</sub> No
    - If Yes, is it covered? .....t905 <sub>1</sub> Yes <sub>3</sub> No
  - b. In a shed? .....t906 <sub>1</sub> Yes <sub>3</sub> No
    - If Yes, is the shed closed?.....t907 <sub>1</sub> Yes <sub>3</sub> No
  
- [If both questions 4a and 4b = No, SKIP to question 7.]**
  
5. What is the minimum distance (in yards) from the on-site litter storage area to the nearest barn?..... t908 \_\_\_\_\_ yards

6. Prior to use, is litter accessible to:
- a. Wild birds? .....t909 <sub>1</sub> Yes <sub>3</sub> No
  - b. Wild animals (e.g., raccoons, opossum, coyotes, foxes)? .....t910 <sub>1</sub> Yes <sub>3</sub> No
  - c. Domestic animals (e.g., dogs, cats)? .....t911 <sub>1</sub> Yes <sub>3</sub> No
7. What was the date that litter was last removed from any barn on this farms?..... t912 \_\_\_\_\_ mm/dd/yy
8. How was litter disposed of most recently?
- a. Composted on-farm .....t913 <sub>1</sub> Yes <sub>3</sub> No  
If Yes, what is the distance (in yards) to the nearest poultry barn?.....t914 \_\_\_\_\_ yards
  - b. Stored on-farm .....t916 <sub>1</sub> Yes <sub>3</sub> No
  - c. Applied to land on this farm.....t917 <sub>1</sub> Yes <sub>3</sub> No  
If Yes, what was the date litter was applied to land?.....t918 \_\_\_\_\_ mm/dd/yy
  - d. Taken off-site.....t919 <sub>1</sub> Yes <sub>3</sub> No
9. Has manure or used litter from other farms been spread on this farm or adjacent farms? ..... t920 <sub>1</sub> Yes <sub>3</sub> No <sub>4</sub> Don't know

**I. DEAD BIRD DISPOSAL**

1. What is the approximate normal daily mortality on this farm? ..... t1001 \_\_\_\_\_ #/day

**Note:** Ratio this number to number of birds in section A, question 2c or 3c (p 4).  
Verify if the mortality is more than 0.01 (1 percent).

2. What are the method(s) of dead bird (daily mortality) disposal on this farm?
- a. Composting .....t1002 <sub>1</sub> Yes <sub>3</sub> No
  - b. Burial .....t1003 <sub>1</sub> Yes <sub>3</sub> No
  - c. Incineration .....t1004 <sub>1</sub> Yes <sub>3</sub> No
  - d. Rendering .....t1005 <sub>1</sub> Yes <sub>3</sub> No
  - e. Landfill .....t1006 <sub>1</sub> Yes <sub>3</sub> No
  - f. Other (specify: \_\_\_\_\_) t1007oth .....t1007 <sub>1</sub> Yes <sub>3</sub> No
3. If question 2a (composting) or question 2b (burial) is Yes, how frequently are carcasses covered with:
- a. Soil? ..... t1008 <sub>1</sub> Daily <sub>2</sub> Every 2 or more days <sub>3</sub> Never
  - b. Manure? ..... t1009 <sub>1</sub> Daily <sub>2</sub> Every 2 or more days <sub>3</sub> Never

4. If question 2d (rendering) is Yes,
- a. Is the carcass bin kept covered? .....t1010      <sub>1</sub> Yes <sub>3</sub> No
  - b. Are carcasses: *[Check one only.]* ..... t1011
    - <sub>1</sub> Taken by the producer/worker to the renderer?
    - <sub>2</sub> Picked up by the renderer from the farm?
  - c. How many times were carcasses moved to the renderer during the 14-day defined risk period? ..... t1012      \_\_\_\_\_ # times
5. What do workers do after handling the carcass bin before returning to the live poultry area? ..... t1013
- \_\_\_\_\_
- \_\_\_\_\_
- \_\_\_\_\_
6. Were any wild birds or wild mammals observed around the dead bird collection area (i.e., burial, compost pile, rendering bin, etc.) during the 14-day defined risk period?
- a. Wild birds .....t1014      <sub>1</sub> Yes <sub>3</sub> No
  - b. Wild mammals.....t1015      <sub>1</sub> Yes <sub>3</sub> No
7. Is there a common collection point (i.e., located off the farm) for dead bird disposal? .....t1016      <sub>1</sub> Yes <sub>3</sub> No
- If Yes, where is the common collection point located? \_\_\_\_\_ t1017

### BARN-LEVEL QUESTIONS

**INSTRUCTIONS:**

- Control farm:** Select one barn to complete this section. **Answer questions for the 14 days prior to the reference date specified on page 4 (the defined risk period).**
- Case farm:** Select the **first** barn on this premises that was confirmed to be HPAI positive. **Answer questions for the 14 days prior to the onset of clinical signs or increased mortality (the defined risk period).**

|  |   |                |
|--|---|----------------|
| 1. What is the barn ID?  |   | t1101          |
| 2. How many barns (flocks) are in the barn structure?  | _____ # barns   |                |
| 3. What separates this barn (flock) from another barn (flock)?   | <input type="checkbox"/> Separate barn structure<br><input type="checkbox"/> Fence<br><input type="checkbox"/> Solid wall | t1102          |
| 4. Are the following type(s) of poultry are present in this barn?  |   |                |
| a. Brooder   | <input type="checkbox"/> Yes <input type="checkbox"/> No  | t1103          |
| b. Grower toms   | <input type="checkbox"/> Yes <input type="checkbox"/> No  | t1104          |
| c. Grower hens   | <input type="checkbox"/> Yes <input type="checkbox"/> No  | t1105          |
| d. Breeders  | <input type="checkbox"/> Yes <input type="checkbox"/> No  | t1106          |
| e. Other   | <input type="checkbox"/> Yes <input type="checkbox"/> No<br>If Yes, specify: _____  | t1107/t1107oth |
| 5. Did birds in this barn have outside access?   | <input type="checkbox"/> Yes <input type="checkbox"/> No  | t1108          |
| 6. How many birds were placed in this barn?  | _____ # birds   | t1109          |
| 7. What was the date of placement in this barn?  | _____ mm/dd/yy  | t1110          |
| 8. How old were birds when placed in this barn?  | _____ days OR _____ weeks   | t1111d/t1111w  |
| 9. Were different stages of production (e.g., brooders and growers) present in this barn at the same time? | <input type="checkbox"/> Yes <input type="checkbox"/> No  | t1112          |
| 10. Was there another health concern in this flock during the defined risk period?                         | <input type="checkbox"/> Yes <input type="checkbox"/> No<br>If Yes, specify condition:<br>_____<br>_____                  | t1113/t1113oth |
| 11. Was this flock being treated for a condition or health concern during the defined risk period?         | <input type="checkbox"/> Yes <input type="checkbox"/> No  | t1114          |
| 12. How old is this barn structure?  | _____ years   | t1115          |
| 13. How long has it been since the last remodel of the barn structure?                                     | _____ years   | t1116          |

|   |  |                        |
|---|--|------------------------|
| <p>14. How well has the barn structure been maintained?<br/>[Enter code 1, 2, or 3.]</p> <p>1. Well<br/>E.g., walls, curtains, and mud boards do not have holes, no visible daylight, the barn is tight and well insulated</p> <p>2. Moderate<br/>E.g., barn could have rust or small holes, mud boards may be damaged, curtains may be torn or not in good repair, curtains may not close all the way, insulation may not be in good repair, the poly may be hanging from the ceiling</p> <p>3. Poor<br/>E.g., holes in walls and mud boards are apparent, tin is rusted, may have leaks in roof, there might be some holes large enough for wild birds to enter, multiple areas with daylight visible, insulation may be hanging from the ceiling</p> | <p>_____ code</p>  | <p>t1117</p>           |
| <p>15. What type of ventilation was used for this barn during the 14 day defined risk period? [Enter Code 1, 2, 3, or 4.]</p> <p>1. Curtain ventilated</p> <p>2. Environmental control/tunnel ventilation</p> <p>3. Side doors (i.e., tip outs)</p> <p>4. Other</p>   | <p>_____ code</p> <p>If 4 (Other), specify:<br/>_____</p>  | <p>t1118/t1118oth</p>  |
| <p>16. If controller information is available, enter the percentage of time that curtains were open and the numbers of days open or partially open during the defined risk period.</p>  | <p>_____ % time</p> <p>_____ # days</p>  | <p>t1119pct/t1119d</p> |
| <p>17. Is intake air filtered?</p>  | <p><input type="checkbox"/><sub>1</sub> Yes <input type="checkbox"/><sub>3</sub> No</p> <p>If Yes, specify type of filter:<br/>_____</p> | <p>t1120/t1120oth</p>  |
| <p>18. What were the minimum and maximum temperatures in the barn during the 14-day defined risk period?</p>  | <p>Minimum: _____</p> <p>Maximum: _____</p>  | <p>t1121/t1122</p>     |
| <p>19. Have you had to repair or replace any feed tank lids in the past 14 days?</p>  | <p><input type="checkbox"/><sub>1</sub> Yes <input type="checkbox"/><sub>3</sub> No</p>  | <p>t1123</p>           |
| <p>20. Have you noticed any feed tank lids open in the past 14 days?</p>  | <p><input type="checkbox"/><sub>1</sub> Yes <input type="checkbox"/><sub>3</sub> No</p>  | <p>t1124</p>           |

|   |   |                       |
|---|---|-----------------------|
| <p>21. Which best describes the ground surface immediately surrounding (within 1 yard) this barn (excluding vehicle approach and loading area). <i>[Enter Code 1, 2, 3, or 4.]</i></p> <p>1. Gravel or hard surface<br/>2. Dirt<br/>3. Short grass<br/>4. Tall grass or brush</p> | <p>_____ code</p>   | <p>t1125</p>          |
| <p>22. Does this barn have a hard-surface entry pad (e.g., concrete, asphalt)?</p>  | <p><input type="checkbox"/><sub>1</sub> Yes <input type="checkbox"/><sub>3</sub> No</p>   | <p>t1126</p>          |
| <p>If Yes,<br/>a. Is the entry pad cleaned and how frequently?</p>  | <p><input type="checkbox"/><sub>1</sub> Yes, <input type="checkbox"/><sub>3</sub> No<br/>If Yes, specify frequency:<br/>_____</p>   | <p>t1127/t1127oth</p> |
| <p>b. Is disinfectant used?</p>   | <p><input type="checkbox"/><sub>1</sub> Yes <input type="checkbox"/><sub>3</sub> No</p>   | <p>t1128</p>          |
| <p>23. How frequently were the following used in this barn during the 14-day defined risk period?</p>   |   |                       |
| <p>a. Locks on the doors</p>  | <p><input type="checkbox"/><sub>1</sub> Used regularly<br/><input type="checkbox"/><sub>2</sub> Not used regularly<br/><input type="checkbox"/><sub>3</sub> Not available</p> | <p>t1129</p>          |
| <p>b. A service room that personnel must enter through that separates “outside area” from “inside area”</p>   | <p><input type="checkbox"/><sub>1</sub> Used regularly<br/><input type="checkbox"/><sub>2</sub> Not used regularly<br/><input type="checkbox"/><sub>3</sub> Not available</p> | <p>t1130</p>          |
| <p>c. Changing area for employees</p>   | <p><input type="checkbox"/><sub>1</sub> Used regularly<br/><input type="checkbox"/><sub>2</sub> Not used regularly<br/><input type="checkbox"/><sub>3</sub> Not available</p> | <p>t1131</p>          |
| <p>d. A shower for employees</p>  | <p><input type="checkbox"/><sub>1</sub> Used regularly<br/><input type="checkbox"/><sub>2</sub> Not used regularly<br/><input type="checkbox"/><sub>3</sub> Not available</p> | <p>t1132</p>          |
| <p>e. Cool cell pads</p>  | <p><input type="checkbox"/><sub>1</sub> Used regularly<br/><input type="checkbox"/><sub>2</sub> Not used regularly<br/><input type="checkbox"/><sub>3</sub> Not available</p> | <p>t1133</p>          |
| <p>f. Mistors</p>   | <p><input type="checkbox"/><sub>1</sub> Used regularly<br/><input type="checkbox"/><sub>2</sub> Not used regularly<br/><input type="checkbox"/><sub>3</sub> Not available</p> | <p>t1134</p>          |
| <p>24. What type of footbath is in use at this barn? <i>[Enter Code 1, 2, 3, or 4.]</i></p> <p>1. Dry (i.e., powdered or particulate)<br/>2. Liquid<br/>3. Other<br/>4. None</p>  | <p>_____ code<br/>If 3 (Other), specify: _____<br/><br/><b>[If 4 (None), SKIP to question 27.]</b></p>  | <p>t1135/t1135oth</p> |
| <p>25. What is the frequency that footbath solutions are changed?</p>   | <p>_____ times/<br/><input type="checkbox"/><sub>1</sub> day, <input type="checkbox"/><sub>2</sub> week, or <input type="checkbox"/><sub>3</sub> month</p>                    | <p>t1136/t1136f</p>   |
| <p>26. What disinfectant is used in the footbaths?</p>  |   | <p>t1137</p>          |

|  |   |                       |
|--|---|-----------------------|
| <p>27. What type(s) of litter is used in this barn?<br/> <i>[Enter Code 1, 2, 3, or 4.]</i></p> <ol style="list-style-type: none"> <li>1. Wood shavings</li> <li>2. Hulls (e.g., oat, rice, sunflower, other)</li> <li>3. Straw</li> <li>4. Other</li> </ol> | <p>_____ code<br/>                 If 4 (Other), specify: _____</p>                       | <p>t1138/t1138oth</p> |
| <p>28. Is the litter bagged (i.e., bailed) or bulk (i.e., load from shavings mill)?</p>  | <p><input type="checkbox"/><sub>1</sub> Bag <input type="checkbox"/><sub>2</sub> Bulk</p> | <p>t1139</p>          |
| <p>29. Who are the supplier(s)/source(s) of litter?</p>  |   | <p>t1140</p>          |
| <p>30. Was litter “tilled” after it was placed in the barn?</p>  | <p><input type="checkbox"/><sub>1</sub> Yes <input type="checkbox"/><sub>3</sub> No</p>   | <p>t1141</p>          |
| <p>If Yes, when was it tilled?</p>   | <p>_____ mm/dd/yy</p>   | <p>t1142</p>          |
| <p>31. How many times was litter added to the barn during the defined risk period?</p>   | <p>_____ times</p>  | <p>t1143</p>          |
| <p>32. What was the date of the last partial clean out in this barn (during the 14-day defined risk period)?</p>   | <p>_____ mm/dd/yy<br/> <input type="checkbox"/><sub>1</sub> No partial cleanout</p>       | <p>t11441/t1144n</p>  |
| <p>If a partial cleanout was done, who performed the partial cleanout?<br/> <i>[Enter Code 1 or 2.]</i></p> <ol style="list-style-type: none"> <li>1. Grower</li> <li>2. Contractor</li> </ol>   | <p>_____ code</p>   | <p>t1145</p>          |
| <p>If contractor, specify name and location:</p>   | <p>Name: _____<br/>                 Location: _____</p>                                   | <p>t1146n/t1146l</p>  |
| <p>33. When was the last full clean out?<br/> <i>[Enter Code 1, 2, or 3.]</i></p> <ol style="list-style-type: none"> <li>1. Prior to this flock</li> <li>2. Two flocks ago</li> <li>3. Three or more flocks ago</li> </ol>                                   | <p>_____ code</p>   | <p>t1147</p>          |
| <p>If a full cleanout was performed, who performed the full cleanout?<br/> <i>[Enter Code 1 or 2.]</i></p> <ol style="list-style-type: none"> <li>1. Grower</li> <li>2. Contractor</li> </ol>  | <p>_____ code</p>   | <p>t1148</p>          |
| <p>If contractor, specify name and location:</p>   | <p>Name: _____<br/>                 Location: _____</p>                                   | <p>t1149n/t1149l</p>  |
| <p>34. Was there a partial load-out while this flock was present?</p>  | <p><input type="checkbox"/><sub>1</sub> Yes <input type="checkbox"/><sub>3</sub> No</p>   | <p>t1150</p>          |
| <p>If Yes, what was the date?</p>  | <p>_____ mm/dd/yy</p>   | <p>t1151</p>          |

|  |  |              |
|--|--|--------------|
| 35. Were the following wild birds seen in this barn during the 14-day defined risk period?           |  |              |
| a. Large birds (e.g., pigeons, crows)  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1152        |
| b. Small birds (e.g., finches, sparrows, starlings)  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1153        |
| 36. How far is this barn (in yards) from:  |  |              |
| a. Dead bird disposal/holding area including carcass bin for rendering?                              | ____ yards   | t1154        |
| b. Litter compost?   | ____ yards<br><input type="checkbox"/> <sub>1</sub> Litter not composted           | t1155/t1155n |
| c. Nearest road?   | ____ yards   | t1156        |
| 37. Did any of the following types of people enter this barn during the 14-day defined risk period?  |  |              |
| a. Federal/State veterinary or animal health worker  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1157        |
| b. Extension agent or university veterinarian  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1158        |
| c. Private or company veterinarian   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1159        |
| d. Company service person  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1160        |
| e. Nutritionist or feed company consultant   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1161        |
| f. Bird delivery personnel   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1162        |
| g. Vaccination crew  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1163        |
| h. Catch crew (bird removal)   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1164        |
| i. Feed delivery personnel   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1165        |
| j. Egg truck personnel (for breeder farms)   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1166        |
| k. Litter delivery services  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1167        |
| l. Litter removal services (e.g., litter broker, litter disposal)                                    | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1168        |
| m. Customer (private individual)   | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1169        |
| n. Wholesaler, buyer, or dealer  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1170        |
| o. Renderer  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1171        |
| p. Occasional worker (e.g., family member, part time help over holiday)                              | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1172        |
| q. Construction workers, repair or maintenance person  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1173        |
| r. Other business visitors (including other producers, meter readers, package delivery (UPS), etc.)  | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1174        |
| s. Other nonbusiness visitors (including neighbors, friends, family members, and school field trips) | <input type="checkbox"/> <sub>1</sub> Yes <input type="checkbox"/> <sub>3</sub> No | t1175        |

## CHECKLIST

### INSTRUCTIONS

This section refers to data that can be acquired through other sources.

1. Please verify grayed areas from the questionnaire.
2. If possible, attach a diagram, farm map, or photographs showing orientation of barn(s), including barn numbers, water location, feed storage, rendering bin, litter storage, ventilation, and windbreaks.
3. If possible, attach photographs of exterior of study barn(s) showing:
  - Exterior structure of the study barn(s), and
  - Ventilation system including exhaust fans and air input and curtains (if present).
4. How many commercial poultry farms (of any production type) are located:
  - a. Within 1 mile of this farm? ..... t1201 \_\_\_\_\_ #
  - b. Within 3 miles of this farm? ..... t1202 \_\_\_\_\_ #
5. How far (in yards or miles) is the nearest backyard flock to this farm? ..... t1203y/t1203m \_\_\_\_\_ yards OR \_\_\_\_\_ miles
6. How far (in yards or miles) is the nearest HPAI-positive premises to this farm? ..... t1204y/t1204m \_\_\_\_\_ yards OR \_\_\_\_\_ miles
7. Collect feed or live haul truck routing information, if available. Determine if trucks are routed to avoid passing positive premises.
8. Collect mortality sheets from both case and control barns.
9. Collect ventilation control information, if available.
10. If available, collect weather data, including historical baselines, relative humidity, wind direction, temperature, and cloud cover.
11. Collect specific age and date of placement for each barn.
12. Which feed mill supplies feed to this farm? \_\_\_\_\_ t1205