

EPIDEMIOLOGIC TRAITS OF AND FACTORS POTENTIALLY CONTRIBUTING TO THE
HIGHLY PATHOGENIC AVIAN INFLUENZA OUTBREAK AMONG COMMERCIAL
POULTRY FARMS IN MINNESOTA AND IOWA BETWEEN APRIL AND JUNE OF 2015

By

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ABSTRACT

EPIDEMIOLOGIC TRAITS OF AND FACTORS POTENTIALLY CONTRIBUTING TO THE HIGHLY PATHOGENIC AVIAN INFLUENZA OUTBREAK AMONG COMMERCIAL POULTRY FARMS IN MINNESOTA AND IOWA BETWEEN APRIL AND JUNE OF 2015

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The largest outbreak of highly pathogenic avian influenza (HPAI) virus among commercial poultry farms in the United States, most notably in Minnesota (MN) and Iowa (IA), took place between December 2014 and June 2015. This study addressed potentially contributing environmental factors through a case-control logistic regression analysis in which environmental traits of MN and IA counties were compared to county infection status. Outcomes were: 0=no infected farms and 1=at least one infected farm in terms of turkey (HPAIT); laying hen and pullet (HPAIC); and turkey, laying hen, and pullet (HPAICOMB) farms. The primary exposure was the percent of total county area which is corn grown for grain. To assess if results were influenced by counties with the greatest number of infected farms, one MN county and two IA counties were removed from the datasets. Prior to removal, significant associations were found between the corn variable and HPAIT for MN, corn variable and HPAIT for IA, corn variable and HPAICOMB for MN, and corn variable and HPAICOMB for IA. After removal, regression coefficient estimates remained approximately the same; these counties did not unduly impact the findings. More consideration might be given to the environmental component of the HPAI virus transmission pathway when attempting to predict spread during an outbreak.

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KEY TO ABBREVIATIONS

adjOR	adjusted odds ratio
OR	odds ratio
CI	confidence interval
USDA	United States Department of Agriculture
APHIS	Animal and Plant Health Inspection Service
HPAI	highly pathogenic avian influenza
LPAI	low pathogenic avian influenza
HPAIT	highly pathogenic avian influenza infection status, where 0=no turkey farms with virus infection and 1=at least one infected turkey farm
HPAIC	highly pathogenic avian influenza infection status, where 0=no laying hen or pullet farms with virus infection and 1=at least one infected laying hen or pullet farm
HPAICOMB	highly pathogenic avian influenza infection status, where 0=no laying hen or pullet or turkey farms with virus infection and 1=at least one infected laying hen or pullet or turkey farm
ALLPOUFA	number of all types of poultry farms per county
TURKFA	number of turkey farms per county
LAYFA	number of laying hen farms per county
PULFA	number of pullet farms per county
LAYPULFA	combined number of laying hen and pullet farms per county
TOTAREAC	acres of total area per county

CROPAC	acres of total cropland per county
CROPPRC	percent of total county area which is cropland
CORNGRAINAC	acres of corn grown for grain per county
CORNGRPRC	percent of total county area which is corn grown for grain
FARMAC	acres of farm land per county
FARMPRC	percent of total county area which is farm land
WATAC	acres of water per county
WATSQPRC	percent of total county area which is water

CHAPTER 1: INTRODUCTION

Between December 2014 and June 2015, roughly 45 million domestic poultry died or were culled due to the highly pathogenic avian influenza (HPAI) virus infection. Culling is a mandatory practice in domestic poultry farms when one or more HPAI-infected birds are detected⁵⁷. The two most affected states, (MN) and Iowa (IA), are ranked highly in agriculture product production and are located within the Mississippi migratory bird route (Figures 13, 14). Asymptomatic wild migratory birds that use farm, particularly corn, fields for shelter and food, have tested positive for HPAI virus⁷⁶. The primary exposure of interest in this study was the percent of total county area which is corn grown for grain (Figures 11, 12). The U.S. Department of Agriculture (USDA) has conducted surveys of domestic poultry operation staff and investigated factors such as wind which may have contributed to the outbreak; however, it has not looked deeply into agricultural and geographic factors. Data on the number of birds on uninfected farms were unavailable; therefore, this study compared the species-specific HPAI virus infection status of MN and IA counties, in terms of infected and uninfected farms per county, to those agricultural and geographic characteristics of the MN and IA counties possibly involved in the mechanism underlying the transmission of HPAI virus from wild migratory birds to domestic birds.

1.1 Objectives

This study aims to summarize the epidemiologic features of the outbreaks in MN and IA and address potential factors which may have contributed to the outbreak by conducting a case-control analysis. contributed to the outbreak by conducting a case-control analysis. contributed to the outbreak by conducting a case-control analysis. contributed to the outbreak by conducting a case-control analysis. contributed to the outbreak by conducting a case-control analysis.

1.2 Hypotheses

1. The greater the percent of total county area which is cropland, the more likely a county is to have at least one infected 1) laying hen or pullet or 2) turkey farm.
2. The greater the percent of total county area which is corn grown for grain, the more likely a county is to have at least one infected 1) laying hen or pullet or 2) turkey farm.
3. The greater the percent of total county area which is farm land, the more likely a county is to have at least one infected 1) laying hen or pullet or 2) turkey farm.
4. The greater the percent of total county area which is water, the more likely a county is to have at least one infected 1) laying hen or pullet or 2) turkey farm.
5. The greater the total number of turkey farms per county, the more likely a county is to have at least one infected turkey farm.
6. The greater the total number of laying hen farms per county, the more likely a county is to have at least one infected laying hen farm.
7. The greater the total number of pullet farms per county, the more likely a county is to have at least one infected pullet farm.

1.3 Hypothesized Transmission Mechanism

MN and IA lie within the Mississippi migratory bird route. Asymptomatic wild migratory birds, particularly water birds, have tested positive for HPAI virus and are suspected to have transmitted the virus to domestic birds during the 2014 to 2015 outbreak⁷⁰. Authors of a 2009 study found that there were more migratory wild birds and more bird species in harvested

sunflower and corn fields than in harvested soybean and small grain fields. Corn field residue “provides considerable vertical dimension” and contains kernels which wild migratory birds find appetizing and relatively easy to eat⁶⁵. In 2015, IA and MN were the first and fourth greatest corn grain harvesting states⁵³. Additionally, northern Iowa contains a wetland region that sees more than half of the North American migratory wild water birds, while around 19 percent of Minnesota is wetland⁶². A 2012 study described several elements possibly involved in the relation of avian influenza virus to wetlands⁶⁷. Wild birds rest in wetland during migration and also utilize it for feeding, breeding, and nesting⁶⁸. MN and IA are also ranked highly in domestic bird production (Figures 13, 14). In the 2015 ranking of states by turkey production, MN and IA are ranked first and eighth, respectively⁸⁰. In the 2016 ranking of states by laying hen inventory, IA is ranked first and MN eighth⁷⁹. IA is also ranked first in the 2014 ranking of states by pullet inventory⁷⁸.

1.4 Biology

HPAI is an A virus that is one of four genera of the Orthomyxoviridae family, which have enveloped virions and a genome with eight single-stranded, negative-sense, ribonucleic acid (RNA) segments⁹. Characteristics of the viral lipoprotein envelope glycoproteins, hemagglutinin and neuraminidase, determine the subtype of the influenza A viruses. Hemagglutinin and neuraminidase antigens give infectious characteristics. Eighteen hemagglutinin subtypes and eleven neuraminidase subtypes are known to exist. All except H17N10 and H18N11, which have been found in bats alone, can infect birds. In common circulation among humans are H1N1 and H3N2. Low-pathogenic avian influenza A (LPAI) can cause mild or no disease in domestic birds. Highly-pathogenic avian influenza A (HPAI) can cause severe disease and mortality of up to ninety to one hundred percent in forty-eight hours in

domestic birds. H5, H7, and H9 viruses can infect both birds and humans. Nine H5 subtypes are known. The majority that are found in wild and domestic birds are LPAI, but HPAI viruses have been found on occasion, and sporadic infection in humans can cause sixty percent mortality^{3,11}. Nine H7 subtypes are known, and the majority that are found in wild and domestic birds are LPAI. Human infection is rare and has occurred in people directly contacting infected birds. LPAI H7 infection in humans can cause mild to moderate illness, while HPAI H7 infection can cause mild to fatal illness. Nine H9 subtypes are known. All which are found in wild and domestic birds are LPAI. Human infection is rare and can cause mild illness³. The main reservoirs of avian influenza are the Anseriformes- and Charadriiformes-order waterfowl, which are ducks, geese, and swans and terns, gulls, and waders, respectively¹².

At the start of virus infection, several hemagglutinin glycoproteins bind to sialic acids on carbohydrate chains of glycolipids and glycoproteins on the surface of cells. After virus replication, neuraminidase removes the sialic acid from the surfaces of infected cells, and new viruses become free to infect other cells. The glycoproteins' characteristics can change to evade the immune response during a pandemic¹⁰. In antigenic drift, mutations gradually alter the hemagglutinin and neuraminidase proteins, in some cases causing previous immune responses to be less effective. In antigenic shift, changes occur quickly when two viruses replicate in one cell and genetic material from both viruses gives rise to a new virion with a new neuraminidase protein, new hemagglutinin protein, or both a new neuraminidase protein and a new hemagglutinin protein. The two viruses can have similar or different animal hosts. If human influenza reassorts with avian influenza, avian influenza can become more transmissible to and among humans and other animals^{7,10}. Antigenic shift can cause the previous immune responses to be completely ineffective⁷.

1.5 Disease Presentation

In domestic birds, signs of HPAI can vary but include sudden death, discharge from the nares, coughing, sneezing, diarrhea, reduced appetite and coordination, swelling or discoloration of the body, and a decline in egg production. HPAI can cause up to 100 percent mortality, commonly in 48 hours, in domestic birds. Transmission among birds usually occurs through contact with airborne secretions and the feces of infected domestic and wild bird³¹. In humans, signs and symptoms of HPAI may include upper respiratory tract illness, myalgia, fever, and conjunctivitis, followed by lower respiratory tract illness, diarrhea, pneumonia, organ failure, encephalitis, and septic shock³². HPAI can cause up to 60 percent mortality in people³³. H5N1 has caused more human cases of disease and death than other avian influenza viruses affecting humans. According to the World Health Organization, there were 258 infections and 154 deaths from H5N1 between 2004 and 2006. Most cases outside the U.S. have occurred in people improperly protected who have directly contacted infected birds or contaminated areas, been within six feet of infected birds, or travelled to a live domestic bird market. The virus is shed in the feces and oral, nasal, and optic fluids of birds³¹. The virus is shed in the feces and oral, nasal, and optic fluids of birds³¹. The virus is shed in the feces and oral, nasal, and optic fluids of birds³¹.

1.6 Early History

Avian influenza was first recorded as “fowl plague” in northern Italy in 1878. It was referred to as Typhus exudatious gallinarum in 1880 and shown to be caused by a filterable virus in 1901. It was classified as a type A influenza virus after the discovery of the “type A influenza virus type-specific ribonucleoprotein” in 1955. In 1981, the description, “HPAI,” was used at the First International Symposium on Avian Influenza in the U.S.¹²

1.7 History in the U.S.

Between 1924 and 1925, wet markets in New York City experienced the first outbreak of HPAI in the U.S. This may have resulted from containers of HPAI virus brought to the U.S. from France for a study in 1923. HPAI spread through infected poultry to New Jersey, Pennsylvania, and Connecticut. HPAI also spread to Indiana, Michigan, West Virginia, Missouri, and Illinois, due in part to contaminated trains. Live poultry travel regulations, quarantining poultry, depopulating farms, cleaning, and disinfecting helped control outbreaks which occurred again in New Jersey in 1929. Wild birds infected with avian influenza virus were discovered using serologic surveys in the U.S., Australia, and Russia in 1968¹².

Pennsylvania chicken farms experienced a notable outbreak of low-pathogenic H5N2 virus in 1983. The same year, Pennsylvania chicken farms suffered an outbreak of a mutated, HPAI variant virus. Over 17 million chickens were culled, indirectly costing about 250 million dollars¹². There was an outbreak of LPAI H7N2 virus on commercial turkey and chicken farms in Virginia in 2002. One of the culling staff developed influenza but survived and did not transmit the virus to other humans. LPAI H7N2 of unknown origin was detected in a person from New York in 2003. The person survived and did not transmit the virus to other humans. An outbreak of HPAI H5N2 virus occurred on a Texas farm with 7,000 chickens in 2004. No humans were affected. Outbreaks of HPAI H5 viruses took place on commercial poultry farms in 21 states between January and June of 2015¹³. Roughly 45 million domestic poultry died from infection or were culled. Economic consequences included export bans on certain U.S. poultry products enforced by about 75 nations^{4,6}. Wild birds were also found to be infected with HPAI viruses between 2014 and 2015. No humans were affected. Commercial turkey farms in Indiana experienced an outbreak of HPAI H7N8 in 2016. No humans were affected. An outbreak of

LPAI H7N2 virus took place among cats in New York City animal shelters in 2016. One person who had contacted the cats became infected but survived and did not transmit the virus to other humans¹³.

The Morbidity and Mortality Weekly Reports (MMWR) on influenza activity from the Centers for Disease Control and Prevention (CDC) over the past 16 years have described emerging influenza strains and outbreaks of avian and swine influenza viruses among domestic animal farms and people. During the 2002 to 2003 influenza season, there was an outbreak of H7N2 virus in Virginia, and 4.7 million turkeys and chickens were culled. One of the culling staff became infected. During the 2003 to 2004 season, there were outbreaks of mostly H7N2 or H7N3 viruses in Delaware, Maryland, New Jersey, Pennsylvania, and Texas. There was an outbreak of H5N2 virus on one Texas poultry farm. During the 2008 to 2009 season, a person in Iowa became infected with swine influenza H1N1 virus after contacting pigs. One person in Texas and one person in South Dakota also became infected with swine influenza virus during the season. During the 2009 to 2010 season, a pandemic swine influenza H1N1 virus infection affected the U.S. The pandemic began in April 2009. There was evidence of transmission of the pandemic strains. Approximately 43 to 89 million people became infected. Additionally, there were three human cases of swine influenza H3N2 infection in Kansas, Iowa, and Minnesota. The person in Minnesota had visited a wet market, and the person in Kansas had contacted pigs. The three people survived. During the 2010 to 2011 season, there were five human cases of swine influenza H3N2 infection, one case in Wisconsin, two cases in Pennsylvania, and two cases in Minnesota. During the 2011 to 2012 season, there were 13 human cases of swine influenza H3N2 infection, two cases in Indiana, three cases in Iowa, two cases in Maine, three cases in Pennsylvania, one case in Utah, and two cases in West Virginia. Additionally, there was one

human case of swine influenza H1N2 infection in Minnesota and one human case of swine influenza H1N1 infection in Wisconsin. During the 2012 to 2013 season, there was one human case of swine influenza H3N2 infection each in Minnesota and Iowa. During the 2013 to 2014 season, there was one human case of swine influenza H3N2 infection in Iowa. During the 2014 to 2015 season, there were three human cases of swine influenza infection, one H3N2 infection in Wisconsin, one H1N1 infection in Minnesota, and one fatal H1N1 infection in Ohio. During the 2015 to 2016 season, there were three human cases of swine influenza infection, one H1N1 infection in Minnesota, one H3N2 infection in New Jersey, and one H1N2 infection in Minnesota¹².

1.8 History in the World

The World Organization for Animal Health Office International des Epizooties (OIE) has been summarizing reports of HPAI by participating countries since 2004. The program began as a response to the outbreak of H5N1 in Southeast Asia at the end of 2003. In 2004, outbreaks of H5N1 were reported by Cambodia, the People's Republic of China, Hong Kong, Indonesia, Japan, the Republic of Korea, Laos, peninsular Malaysia, Thailand, and Vietnam. H5N2 was reported by Chinese Taipei²¹.

In 2005, the OIE received reports of H5N1 from Cambodia, the People's Republic of China, Croatia, Hong Kong, Indonesia, Kazakhstan, peninsular Malaysia, Mongolia, Romania, Russia, Thailand, Turkey, Ukraine, and Vietnam. H5N2 was reported by Zimbabwe. H5 was reported by the Philippines. H7N7 was reported by the Democratic People's Republic of Korea²¹.

In 2006, H5N1 was reported by Afghanistan, Albania, Austria, Azerbaijan, Bosnia and Herzegovina, Bulgaria, Burkina Faso, Cambodia, Cameroon, the People's Republic of China,

Croatia, the Czech Republic, Cote d'Ivoire, Denmark, Djibouti, Egypt, France, Georgia, Germany, Greece, Hong Kong, Hungary, India, Indonesia, Iran, Iraq, Israel, Italy, Jordan, Kazakhstan, the Democratic People's Republic of Korea, the Republic of Korea, Laos, peninsular Malaysia, Mongolia, Myanmar, Niger, Nigeria, Pakistan, Palestinian Autonomous Territories, Poland, Romania, Russia, Serbia and Montenegro, Slovakia, Slovenia, Spain, Sudan, Sweden, Switzerland, Thailand, Turkey, Ukraine, the United Kingdom, and Vietnam. H5N2 was reported by Zimbabwe and South Africa²¹.

In 2007, the OIE saw reports of H5N1 by Afghanistan, Bangladesh, Benin, Cambodia, the People's Republic of China, the Czech Republic, Cote d'Ivoire, Djibouti, Egypt, France, Germany, Ghana, Hong Kong, Hungary, India, Japan, the Republic of Korea, Kuwait, Laos, peninsular Malaysia, Myanmar, Pakistan, Poland, Romania, Russia, Saudi Arabia, Slovenia, Sudan, Thailand, Togo, Turkey, Ukraine, the United Kingdom, and Vietnam. Canada reported H7N3²¹.

In 2008, reports of H5N1 were made by Bangladesh, Benin, Cambodia, the People's Republic of China, Egypt, Germany, Hong Kong, India, Iran, Israel, Japan, the Republic of Korea, Laos, Myanmar, Nigeria, Pakistan, Poland, Romania, Russia, Saudi Arabia, Switzerland, Thailand, Togo, Turkey, Ukraine, the United Kingdom, and Vietnam. The United Kingdom also reported H7N7. Canada reported H7N3²¹.

In 2009, H5N1 was reported by Afghanistan, Bangladesh, Cambodia, the People's Republic of China, Germany, Hong Kong, India, Japan, Laos, Mongolia, Nepal, Nigeria, Russia, Thailand, Togo, and Vietnam. Spain reported H7N7²¹.

In 2010, reports of H5N1 were made by Bangladesh, Bhutan, Bulgaria, Cambodia, the People's Republic of China, Hong Kong, India, Israel, Japan, the Republic of Korea, Laos, Myanmar, Nepal, Romania, Russia, and Vietnam. Spain reported H7N7. Mongolia reported H5¹⁷.

In 2011, H5N1 was reported by Bangladesh, Cambodia, the People's Republic of China, Hong Kong, India, Indonesia, Iran, Israel, Japan, the Republic of Korea, Mongolia, Myanmar, Nepal, Palestinian Autonomous Territories, and Vietnam. H5N2 was reported by South Africa²¹.

In 2012, H5N1 was reported by Bangladesh, Bhutan, Cambodia, the People's Republic of China, Chinese Taipei, Hong Kong, India, Iran, Israel, Myanmar, Nepal, and Vietnam. Australia reported H7N7. Chinese Taipei and South Africa reported H5N2. Israel reported H5. Mexico reported H7N3²¹.

In 2013, H5N1 was reported by Bangladesh, Bhutan, Cambodia, the People's Republic of China, Hong Kong, India, the Democratic People's Republic of Korea, Nepal, and Vietnam. H7N7 was reported by Australia and Italy. H7N2 was reported by Australia. H5N2 was reported by the People's Republic of China, Chinese Taipei, and South Africa. H7N3 was reported by Mexico²¹.

In 2014, H5N1 was reported by Cambodia, the People's Republic of China, India, the Democratic People's Republic of Korea, Libya, Nepal, Russia, and Vietnam. H7N2 was reported by Australia. H5N2 was reported by Canada, the People's Republic of China, the U.S. of America, Chinese Taipei, and the U.S. of America. H5N8 was reported by Germany, Italy, Japan, the Republic of Korea, the Netherlands, Russia, the United Kingdom, the U.S. of America, and the People's Republic of China. H5N6 was reported by Laos, Vietnam, and the

People's Republic of China. H7N3 was reported by Mexico. H5N3 was reported by the People's Republic of China²¹.

In 2015, H5N1 was reported by Bhutan, Bulgaria, Burkina Faso, Cambodia, Canada, the People's Republic of China, Cote d'Ivoire, France, Ghana, India, Iran, Israel, Kazakhstan, Libya, Myanmar, Niger, Nigeria, the Palestinian Autonomous Territories, Romania, Russia, Turkey, the U.S. of America, and Vietnam. H5N2 was reported by Canada, the People's Republic of China, Chinese Taipei, France, and the U.S. of America. H5N8 was reported by Canada, Chinese Taipei, Germany, Hungary, Italy, Japan, the Republic of Korea, the Netherlands, Russia, Sweden, the United Kingdom, and the U.S. of America. H5N6 was reported by the People's Republic of China, Hong Kong, Laos, and Vietnam. H5N3 was reported by Chinese Taipei. H5N9 was reported by France. H7N7 was reported by Germany and the United Kingdom. H7N3 was reported by Mexico. H5 was reported by the Palestinian Autonomous Territories²¹.

In 2016, H5N1 was reported by Bangladesh, Bhutan, Burkina Faso, Cambodia, Cameroon, the People's Republic of China, Cote d'Ivoire, France, Ghana, India, Iraq, Laos, Lebanon, Myanmar, Niger, Nigeria, Togo, and Vietnam. H5N6 was reported by Vietnam, the Republic of Korea, Japan, Hong Kong, and the People's Republic of China. H5N2 was reported by the U.S. of America, France, Chinese Taipei, and the People's Republic of China. H7N8 was reported by the U.S. of America. H5N8 was reported by the United Kingdom, Tunisia, Switzerland, Sweden, Serbia, Russia, Romania, Poland, Nigeria, Netherlands, the Republic of Korea, Israel, Iran, India, Hungary, Greece, Germany, France, Finland, Egypt, Denmark, Croatia, Chinese Taipei, Canada, and Austria. H5 was reported by Ukraine, Tunisia, Russian Palestinian Autonomous Territories, and Bulgaria. H5N5 was reported by the Netherlands and Montenegro.

H7N3 was reported by Mexico. H7N7 was reported by Italy. H5N9 was reported by France. H5N3 was reported by Chinese Taipei. H7N1 was reported by Algeria²¹.

1.9 Events Pertinent to the 2014/2015 HPAI Outbreak in Minnesota and Iowa

1.9.1 Asia

An outbreak of HPAI H5N8 virus infection occurred among poultry in South Korea and Japan between January and April of 2014. In China, outbreaks of HPAI H5N1, H5N2, H5N6, and H5N8 virus infection followed in September. South Korea had additional cases of H5N8 virus infection among its commercial poultry in late September. In November, cases of H5N8 virus infection were seen in Japan. Taiwan had outbreaks of H5N2, H5N3, and H5N8 virus infection in early 2015⁵⁶.

1.9.2 Europe

In Germany, H5N8 virus was detected in commercial poultry and in a wild duck in early November 2014. The United Kingdom, The Netherlands, and Italy detected H5N8 virus in commercial poultry in mid-December⁵⁶.

1.9.3 North America

Wild waterfowl using the East Asia / Australia migration route probably brought the HPAI H5N8 virus to North America. A bird in North America became infected with both the HPAI H5N8 virus and an endemic LPAI virus, and genetic reassortment occurred to bring about the H5N1 and H5N2 viruses. British Columbia, Canada detected H5N2 virus in its commercial poultry in late November 2014. H5N8 virus was detected in captive wild birds in Washington state in early December. Oregon found H5N8 virus in backyard poultry in December. In late December, Washington state detected H5N1 virus in a wild duck. California had commercial

turkeys infected with H5N8 virus in mid-January 2015. In March 2015, H5N2 virus was detected in poultry in Arkansas, Kansas, Minnesota, and Missouri⁵⁶.

There were over 200 confirmed detections of HPAI A in wild birds and domestic poultry between December 2014 and May 2015 in the U.S., with over 40 million birds in 20 states exposed or infected²⁷. Domestic and wild birds in the U.S. were affected by the subtypes H5N1, H5N2, and H5N8, the majority of domestic birds being affected by H5N2. By June 2015, Minnesota had the greatest number of cases followed by Iowa. Birds on all infected farms were culled. Throughout the U.S., turkeys were affected most followed by laying hens, mixed poultry, pullets, chickens in general, and, with an equal number of cases, ducks, mixed game fowl, and pheasants. After January 2016, one report of LPAI H7N8 virus infection came from a turkey farm in Indiana, where 43,000 turkeys were culled³⁰.

1.10 Study Justification

Consequences of the outbreak included lost jobs and export bans on certain U.S. poultry products. The prices of poultry products in the U.S. increased between December 2014 and the spring of 2015, and about 75 nations restricted or banned the import of these products^{4,6}. By August 2015, 8,444 jobs and therefore 427 million dollars in wages and 145 million dollars in taxes were lost in Iowa. Veterinarians, suppliers of poultry feed, truck transporters, poultry processors, and others suffered negative economic effects⁵. The government granted indemnities to owners of affected farms⁸². Understanding what may have contributed to the outbreak is necessary to protect the U.S. agricultural industry, domestic bird welfare, and human health. As MN and IA have large poultry and swine inventories, and influenza tends to undergo antigenic changes when passing between hosts, there is no assurance that HPAI will not become more transmissible to and among humans in the future.

CHAPTER 2: METHODS

This study was deemed as exempt from approval by the Michigan State University Institutional Review Board (IRB).

2.1 Study Population and Design

The USDA Animal and Plant Health Inspection Service (APHIS) outbreak dataset provided data on the infected commercial laying hen, turkey, and pullet farms US states and counties⁷⁴. Since information was inconsistently available for the number of farms that were not infected for IA and MN, the analysis had to be performed at the county level. Therefore, 186 counties were included in the analytic sample at the county level. This study compared the species-specific HPAI virus infection status of MN and IA counties to those agricultural and geographic characteristics of the MN and IA counties possibly involved in the mechanism underlying the transmission of HPAI virus from wild migratory birds to domestic birds. The 2012 USDA Census of Agriculture provided information on the agricultural traits of the counties, while the 2010 U.S. Census provided information on the geographic traits^{72,73}.

2.2 Measures

The 2012 USDA Census of Agriculture provided information on the percent of total county area which is cropland, the percent of total county area which is corn grown for grain, the percent of total county area which is farm land, the total number of turkey farms per county, the total number of laying hen farms per county, and the total number of pullet farms per county⁷². The 2010 U.S. Census provided information on the percent of total county area which is water⁷³. The USDA APHIS outbreak dataset provided information on the HPAI virus infection status of the counties⁷⁴. Measurement methods for the 2012 USDA Census of Agriculture are described in the Census Appendix B⁵². It defines a farm as an operation which produces or sells greater than

or equal to \$1000 of agricultural products. Measurement methods for the 2010 U.S. Census are described in the technical assessment supplement⁷⁵. The USDA APHIS outbreak data were collected in part via survey of farm personnel and on-farm sampling⁷¹. Data from the 2012 USDA Census of Agriculture were arranged in a PDF file. For the analysis, the data were manually imported into a Microsoft Excel spreadsheet in comma delimited values format. Data from the 2010 U.S. Census were arranged in a Microsoft Excel file. They were copied and pasted into the comma delimited value file. Data from the USDA APHIS outbreak dataset were arranged in a PDF file and manually imported into the comma delimited value file.

2.3 Analysis

Logistic regression was performed with three dichotomous outcomes of interest and continuous exposure variables using SAS version 9.4 (SAS Institute, Inc., Cary, NC). Significance was considered at $p < 0.05$ level.

Original exposures were the acres of total cropland per county, the acres of corn grown for grain per county, the acres of farm land per county, the square miles of water per county, the total number of turkey farms per county, the total number of laying hen farms per county, and the total number of pullet farms per county. With the exception of the farm numbers and the square miles of water per county, exposures were converted to square miles. Next, all exposures but the farm numbers were calculated as proportions of the total county area. Finally, percents of the total county area were created for all exposures but the farm numbers, and the number of laying hen and pullet farms per county was combined. Pullets are simply young laying hens, and the individual number of laying hen and pullet farms per county was small. Final exposures were the percent of total county area which is cropland (CROPPRC), the percent of total county area which is corn grown for grain (CORNGRPC), the percent of total county area which is farm

land (FARMPRC), the percent of total county area which is water (WATSQPRC), the total number of turkey farms per county (TURKFA), and the combined number of laying hen and pullet farms per county (LAYPULFA).

Two species-specific dichotomous outcomes and one unspecific dichotomous outcome were used in the analysis. They were HPAIC, where 0=no laying hen or pullet farms with virus infection and 1=at least one infected laying hen or pullet farm; HPAIT, where 0=no turkey farms with virus infection and 1=at least one infected turkey farm; and HPAICOMB, where 0=no laying hen or pullet or turkey farms with virus infection and 1=at least one infected laying hen or pullet or turkey farm.

To assess if the counties with the greatest number of infected farms influenced the regression coefficient estimates, one county from the MN dataset (Kandiyohi) and two counties from the IA dataset (Buena Vista, Sioux) were removed and logistic regression analyses with these counties were compared to those without these counties. These counties were infected early and frequently during the observation period.

CHAPTER 3: RESULTS

Table 1a shows the number of infected farms for each state and for IA and MN on a county level. Table 1b shows the characteristics of the cases on the farm level. Data on the characteristics were provided by the USDA APHIS outbreak dataset. Commercial domestic poultry operations in MN and IA were selected for this study. Within the original outbreak dataset (N=232), which consisted of infected farms, 77 infected IA farms made up 33.2 percent of the sample, while 110 infected MN farms made up 47.4 percent of the sample. After applying inclusion and exclusion criteria, no MN or IA counties were excluded based on the commercial operation criterion; counties with excluded “backyard” operations still contained infected commercial operations. 71 infected IA farms made up 39.4 percent of the analytic sample, while 109 infected MN farms made up 60.6 percent of the analytic sample. Within IA, Buena Vista County, with 15 infected farms, made up the largest percentage, 21.1 percent, of the analytic sample (Figure 8). Sioux County (14 infected farms, 19.7 percent) and Sac County (8 infected farms, 11.3 percent) followed. Within MN, Kandiyohi County, with 40 infected farms, made up the largest percentage, 36.7 percent, of the analytic sample (Figure 7). Stearns County (14 infected farms, 12.8 percent) and Renville County (8 infected farms, 7.3 percent) followed. Considering MN and IA together, infected farms with between zero and 100,000 birds, 135 infected farms, made up the largest percentage, 75.8 percent, of the analytic sample. Infected farms with greater than 500,000 birds (16 infected farms, 9.0 percent) and infected farms with between greater than 100,000 birds and 200,000 birds (11 infected farms, 6.2 percent) followed. Considering MN and IA together, turkey farms, 139 infected farms, made up the largest percentage, 77.2 percent, of the analytic sample. Laying hen farms (27 infected farms, 15 percent) and pullet farms (14 infected farms, 7.8 percent) followed. Considering MN and IA

together, the majority of infected farms (173 infected farms, 96.1 percent) were infected with HPAI H5N2 virus in the analytic sample. The unspecific “Dangerous Contact Highly Pathogenic” (6 infected farms, 3.3 percent) and HPAI H5 (one infected farm, 0.6 percent) categories followed.

Figure 1 shows the derivation of the analytic sample on the farm level through the application of inclusion and exclusion criteria. Farms in states other than MN and IA (N=45) were first excluded from the analytic sample. Farms that are “backyard,” rather than commercial, operations (N=7) were next excluded for MN and IA to give a count of 180 farms for the infected farms analytic sample.

Figures 2 and 3 show the full outbreak for descriptive purposes (the outbreak took place between 4/13/2015 and 6/16/2015 in IA and between 3/5/2015 and 6/4/2015 in MN); however, in order to compare the infection rates in the two states, outbreak dates were restricted to April 2015 through June of 2015 in the analysis.

Figure 4 shows that the derivation of counties to be considered in the county analytic sample. MN has 87 counties, while IA has 99 counties. Two MN counties were missing information on the acres of corn grown for grain per county, which was used to create the percent of total county area which is corn grown for grain variable. These counties were not excluded since the percent of total county area which is corn grown for grain was only one of the exposures of interest. After applying inclusion and exclusion criteria, no MN or IA counties were excluded based on the commercial operation criterion; counties with excluded “backyard” operations still contained infected commercial operations.

Table 2 illustrates the geographical characteristics of the MN and IA counties in the analytic sample (N=186). The 2012 USDA Census of Agriculture and the 2010 U.S. Census were used to determine the characteristics. The percentage of infected counties for the two states differed with 18.2 percent for IA and 26.4 percent for MN. Compared to infected IA counties, infected MN counties had a greater mean value for the acres of total area per county, greater mean value for the acres of water per county, smaller mean value for the acres of farm land per county, smaller mean value for the acres of total cropland per county, smaller mean value for the acres of corn grown for grain per county, greater mean value for the number of all types of poultry farms per county, greater mean value for the number of laying hen farms per county, greater mean value for the number of turkey farms per county, and greater mean value for the number of pullet farms per county.

Considering laying hens, pullets, and turkeys, there were 64 HPAI-uninfected MN counties and 23 HPAI-infected MN counties (Figure 5). Table 2 shows that the infected MN counties relative to the uninfected had a smaller mean value for the acres of total area per county, smaller mean value for the acres of water per county, greater mean value for the acres of farm land per county, greater mean value for the acres of total cropland per county, greater mean value for the acres of corn grown for grain per county, roughly equal mean value for the number of all types of poultry farms per county, smaller mean value for the number of laying hen farms per county, greater mean value for the number of turkey farms per county, and smaller mean value for the number of pullet farms per county.

There were 81 HPAI-uninfected IA counties and 18 HPAI-infected IA counties (Figure 6). Table 2 shows that the infected IA counties relative to the uninfected had a greater mean value for the acres of total area per county, smaller mean value for the acres of water per county,

greater mean value for the acres of farm land per county, greater mean value for the acres of total cropland per county, greater mean value for the acres of corn grown for grain per county, smaller mean value for the number of all types of poultry farms per county, smaller mean value for the number of laying hen farms per county, greater mean value for the number of turkey farms per county, and smaller mean value for the number of pullet farms per county.

Table 4 summarizes the HPAI outbreak in MN and IA between April and June of 2015. Within IA, the greatest incidence rate per 100,000 farm-days of observation was seen among turkey farms, followed by pullet farms and laying hen farms. Within MN, the greatest incidence rate was also seen among turkey farms, followed by pullet and laying hen farms. Compared to IA, MN had a greater incidence rate in terms of turkey farms, a smaller incidence rate in terms of pullet farms, and a smaller incidence rate in terms of laying hen farms. The MN/IA incidence rate ratio was greatest for turkey farms, followed by laying hen and pullet farms.

Table 4 and Figure 9 shows that, overall, the greatest cumulative incidence (CI) of virus infection was seen in turkey farms, followed by pullet and laying hen farms. The same was true for incidence density (ID). State specific: 1) MN turkey farms had greater CI and ID values than IA turkey farms; 2) IA laying hen farms had greater CI and ID values than MN laying hen farms; and 3) IA pullet farms had greater CI and ID values than MN pullet farms. The MN/IA incidence rate ratio for turkey farms was 2.1 (95 percent confidence interval: 1.4-3.1), for laying hen farms was 0.2 (0.05-0.5), and for pullet farms was 0.07 (0.009-0.5).

Table 5a shows the results of the logistic regression analysis with the HPAIC outcome, where 0=no laying hen or pullet farms with HPAI virus infection and 1=at least one infected laying hen or pullet farm, based on data from all counties. In the univariate analysis for MN, the percent of total county area which is farm land was significant at $p < 0.05$ and the percent of total

county area which is cropland was significant at $p < 0.1$. The odds ratio estimates were 7.8 and 2.6, respectively. In the univariate analysis for IA, the percent of total county area which is cropland and the percent of total county area which is farm land were significant at $p < 0.05$. The odds ratio estimates were 1.9 and 2.5, respectively. In the multivariate analysis for MN, no exposures remained significant after adjustment for the other exposures in the model due to high correlation between exposures. The same was true for IA.

Table 5aa shows the results of the logistic regression analysis following the removal of the counties with the greatest number of infected farms: Kandiyohi (MN), Buena Vista (IA), and Sioux (IA). The HPAIC outcome, where 0=no laying hen or pullet farms with virus infection and 1=at least one infected laying hen or pullet farm, is used. In the univariate analysis for MN, the percent of total county area which is cropland was significant at $p < 0.1$, and the percent of total county area which is farm land was significant at $p < 0.05$. In the univariate analysis for IA, the percent of total county area which is cropland and the percent of total county area which is farm land were significant at $p < 0.1$. In the multivariate analysis for MN, no exposures remained significant after adjustment for the other exposures in the model due to high correlation between exposures. The same was true for IA.

Table 5b shows the results of the logistic regression analysis with the HPAIT outcome, where 0=no turkey farms with HPAI virus infection and 1=at least one infected turkey farm, based on data from all counties. In the univariate analysis for MN, the number of turkey farms per county was significant at $p < 0.05$; the percent of total county area which is cropland, the percent of total county area which is corn, and the percent of total county area which is farm land were significant at $p < 0.01$; and the percent of total county area which is water was significant at $p < 0.1$. In the univariate analysis for IA, the number of turkey farms per county was significant at

$p < 0.01$, and the combined number of laying hen and pullet farms per county, the percent of total county area which is cropland, the corn variable, and the percent of total county area which is farm land were significant at $p < 0.05$.

In the multivariate analysis for MN, for the outcome HPAIT (Table 5b), during the model building process, only two exposures retained their statistical significance when adjusted for each other: the number of turkey farms per county at $p < 0.05$ and the corn variable at $p < 0.01$. The odds ratio estimates were 1.1 and 2.0, respectively. In the multivariate analysis for IA, the model building process yielded three models where mutually adjusted exposures remained significant: 1) the number of turkey farms per county, the corn variable; 2) the combined number of laying hen and pullet farms per county, the corn variable; and 3) the number of turkey farms per county, the combined number of laying hen and pullet farms per county, the corn variable. Estimates were significant 1) at $p < 0.01$ for the number of turkey farms per county and at $p < 0.05$ for the corn variable; 2) at $p < 0.05$ for the combined number of laying hen and pullet farms per county and the corn variable; and 3) at $p < 0.05$ for the number of turkey farms per county and the combined number of laying hen and pullet farms per county and at $p < 0.1$ for the corn variable. The odds ratio estimates were 1) 1.3 and 3.3; 2) 0.9 and 5.1; and 3) 1.4, 0.9, and 4.1, respectively.

Table 5bb shows the results of the logistic regression analysis following the removal of the counties with the greatest number of infected farms: Kandiyohi (MN), Buena Vista (IA), and Sioux (IA). The HPAIT outcome, where 0=no laying hen or pullet farms with HPAI virus infection and 1=at least one infected laying hen or pullet farm, is used. In the univariate analysis for MN, the percent of total county area which is water was significant at $p < 0.1$; the number of turkey farms per county was significant at $p < 0.05$; and the percent of total county area which is

cropland, the percent of total county area which is corn grown for grain, and the percent of total county area which is farm land were significant at $p<0.01$. In the univariate analysis for IA, the combined number of laying hen and pullet farms per county and the percent of total county area which is farm land were significant at $p<0.1$. The number of turkey farms per county, the percent of total county area which is cropland, and the percent of total county area which is corn grown for grain were significant at $p<0.05$. In the multivariate analysis for MN, a model with the number of turkey farms per county and the percent of total county area which is corn grown for grain was possible. Estimates were significant at $p<0.05$ for the number of turkey farms per county and at $p<0.01$ for the percent of total county area which is corn grown for grain. Odds ratio estimates were 1.1 and 2.0, respectively.

Table 5c shows the results of the logistic regression analysis with the HPAICOMB outcome, where 0=no laying hen or pullet or turkey farms with HPAI virus infection and 1=at least one infected laying hen or pullet or turkey farm, based on data from all counties. In the univariate analysis for MN, the number of turkey farms per county and the percent of total county area which is water were significant at $p<0.1$; the percent of total county area which is cropland and the percent of total county area which is farm land were significant at $p<0.01$; and the corn variable was significant at $p<0.001$. In the univariate analysis for IA, the combined number of laying hen and pullet farms per county and the percent of total county area which is water were significant at $p<0.1$; the percent of total county area which is cropland and the percent of total county area which is farm land were significant at $p<0.01$; and the corn variable was significant at $p<0.05$.

In the multivariate analysis for MN, for the outcome HPAICOMB (Table 5c), during the model building process, only two exposures retained their statistical significance when adjusted

for each other: the number of turkey farms per county at $p < 0.05$ and the corn variable at $p < 0.001$. The odds ratio estimates were 1.1 and 2.1, respectively. In the multivariate analysis for IA, two exposures retained their statistical significance when adjusted for each other: the combined number of laying hen and pullet farms per county at $p < 0.1$ and the corn variable at $p < 0.05$. The odds ratio estimates were 1.0 and 1.8, respectively.

Table 5cc shows the results of the logistic regression analysis following the removal of the counties with the greatest number of infected farms: Kandiyohi (MN), Buena Vista (IA), and Sioux (IA). The HPAICOMB outcome, where 0=no laying hen or pullet or turkey farms with HPAI virus infection and 1=at least one infected laying hen or pullet or turkey farm, is used. In the univariate analysis for MN, the number of turkey farms per county and the percent of total county area which is water were significant at $p < 0.1$; the percent of total county area which is cropland and the percent of total county area which is farm land were significant at $p < 0.01$; and the percent of total county area which is corn grown for grain was significant at $p < 0.001$. In the univariate analysis for IA, the percent of total county area which is water was significant at $p < 0.1$; the combined number of laying hen and pullet farms per county and the percent of total county area which is corn grown for grain were significant at $p < 0.05$; and the percent of total county area which is cropland and the percent of total county area which is farm land were significant at $p < 0.01$.

In the multivariate analysis for MN, for the outcome HPAICOMB (Table 5cc), during the model building process, only two exposures retained their statistical significance when adjusted for each other: the number of turkey farms per county at $p < 0.1$ and the percent of total county area which is corn grown for grain at $p < 0.001$. Odds ratio estimates were 1.1 and 2.1, respectively. In the multivariate analysis for IA, only two exposures retained their statistical

significance when adjusted for each other: the combined number of laying hen and pullet farms per county at $p < 0.05$ and the percent of total county area which is corn grown for grain at $p < 0.05$. Odds ratio estimates were 1.0 and 1.8, respectively.

A review of the univariate and multivariate analyses showed that the corn variable is the main exposure which was most consistently statistically significant for all three outcome variables. Following is a summary of the results for the corn variable as observed in Tables 5a through 5cc. Table 5a shows that, in the unadjusted model for MN and the HPAIC outcome, for a 10 percent increase in the percent of total county area which is corn, the odds that a county has at least one infected laying hen or pullet farm versus having no laying hen or pullet farms with virus infection increase by a factor of two. This is not significant but shows a trend toward significance. After the counties with the greatest number of infected farms were removed in Table 5aa, unadjusted, every 10 percent increase in the corn variable is associated with a 101 percent increase in the odds that a county has at least one infected laying hen or pullet farm. Again, this shows a trend toward significance but is not significant. In the unadjusted model for IA and HPAIC as shown in Table 5a, for every 10 percent increase in the corn variable, the odds that a county has at least one infected laying hen or pullet farm versus having no laying hen or pullet farms with virus infection increase by a factor of 1.4. This is not significant. After the aforementioned counties were removed in Table 5aa, unadjusted, every 10 percent increase in the corn variable is associated with a 34 percent increase in the odds that a county has at least one infected laying hen or pullet farm. This is not significant.

In the unadjusted model for MN and HPAIT in Table 5b, for every 10 percent increase corn, the odds that a county has at least one infected turkey farm versus having no turkey farms with virus infection increase by a factor of 1.8. This is significant. Adjusting for the number of

turkey farms per county, for every 10 percent increase in corn, holding the number of turkey farms per county fixed, the odds that a county has at least one infected turkey farm versus having no turkey farms with virus infection increase by a factor of two. Significance has increased with adjustment. After the aforementioned counties were removed in Table 5bb, unadjusted, every 10 percent increase in corn is associated with an 82 percent increase in the odds that a county has at least one infected turkey farm. This is significant. Adjusting for the number of turkey farms per county, every 10 percent increase in corn is associated with a 99 percent increase in the odds that a county has at least one infected turkey farm. The significance remains approximately the same with adjustment. In the unadjusted model for IA and HPAIT as shown in Table 5b, for every 10 percent increase in corn, the odds that a county has at least one infected turkey farm versus having no turkey farms with virus infection increase by a factor of 3.2. This is significant. Adjusting for the number of turkey farms per county, for every 10 percent increase in corn, holding the number of turkey farms per county fixed, the odds that a county has at least one infected turkey farm versus having no turkey farms with virus infection increase by a factor of 3.3. This coefficient for corn becomes less significant with this adjustment. Adjusting for the combined number of laying hen and pullet farms per county, for every 10 percent increase in corn, holding the combined number of laying hen and pullet farms per county fixed, the odds that a county has at least one infected turkey farm versus having no turkey farms with virus infection increase by a factor of 5.1. The significance remains approximately the same. Adjusting for the number of turkey farms per county and the combined number of laying hen and pullet farms per county, for every 10 percent increase in corn, holding the number of turkey farms per county and the combined number of laying hen and pullet farms per county fixed, the odds that a county has at least one infected turkey farm versus having no turkey farms with virus infection increase by a

factor of 4.1. Significance of the coefficient for corn decreases with this adjustment. After the counties were removed in Table 5bb, unadjusted, every 10 percent increase in corn is associated with a 223 percent increase in the odds that a county has at least one infected turkey farm. This is significant.

Table 5c shows that, in the unadjusted model for MN and HPAICOMB, for every 10 percent increase in corn, the odds that a county has at least one infected laying hen or pullet or turkey farm versus having no laying hen or pullet or turkey farms with virus infection increase by a factor of 1.9. This is highly significant. Adjusting for the number of turkey farms per county, for every 10 percent increase in corn, holding the number of turkey farms per county fixed, the odds that a county has at least one infected laying hen or pullet or turkey farm versus having no laying hen or pullet or turkey farms with virus infection increase by a factor of 2.1. Significance decreases slightly with this adjustment. After the aforementioned counties were removed in Table 5cc, unadjusted, every 10 percent increase in corn is associated with a 92 percent increase in the odds that a county has at least one infected laying hen or pullet or turkey farm. This is highly significant. Adjusting for the number of turkey farms per county, every 10 percent increase in corn is associated with a 105 percent increase in the odds that a county has at least one infected laying hen or pullet or turkey farm. Significance decreases slightly with this adjustment. As shown in Table 5c, in the unadjusted model for IA and HPAICOMB, for every 10 percent increase in corn, the odds that a county has at least one infected laying hen or pullet or turkey farm versus having no laying hen or pullet or turkey farms with virus infection increase by a factor of 1.9. This is significant. Adjusting for the combined number of laying hen and pullet farms per county, for every 10 percent increase in corn, holding the combined number of laying hen and pullet farms per county fixed, the odds that a county has at least one infected

laying hen or pullet or turkey farm versus having no laying hen or pullet or turkey farms with virus infection increase by a factor of 1.8. Significance remains unchanged. After the counties were removed in Table 5cc, unadjusted, every 10 percent increase in corn is associated with an 81 percent increase in the odds that a county has at least one infected laying hen or pullet or turkey farm. This is significant. Adjusting for the combined number of laying hen and pullet farms per county, every 10 percent increase in corn is associated with an 80 percent increase in the odds that a county has at least one infected laying hen or pullet or turkey farm. Significance remains unchanged.

The first three counties to be infected in MN were Pope, Lac Qui Parle, and Stearns (Figure 10). Pope is ranked 34th out of 87 counties for the acres of corn grown for grain per county, while Lac Qui Parle is ranked 10th, and Stearns is ranked fifth. For the square miles of water per county, Pope is ranked 19th, Lac Qui Parle 53rd, and Stearns 20th. For the number of turkey farms per county, Pope is ranked 44th, Lac Qui Parle 71st, and Stearns first. For the number of laying hen farms per county, Pope is ranked 55th, Lac Qui Parle 77th, and Stearns first. Finally, for the number of pullet farms per county, Pope is ranked 43rd, Lac Qui Parle 60th, and Stearns 15th. Respectively, Pope, Lac Qui Parle, and Stearns accounted for about two, one, and 13 percent of all HPAI virus-infected commercial poultry farms in MN.

The first three counties to be infected in IA were Buena Vista, Osceola, and Sac. Buena Vista is ranked 28th out of 99 counties for the acres of corn grown for grain per county, while Osceola is ranked 67th, and Sac is ranked 25th. For the square miles of water per county, Buena Vista is ranked 25th, Osceola 80th, and Sac 34th. For the number of turkey farms per county, Buena Vista is ranked first, Osceola 38th, and Sac second. For the number of laying hen farms per county, Buena Vista is ranked 97th, Osceola 90th, and Sac 88th. Finally, for the number of

pullet farms per county, Buena Vista is ranked 53rd, Osceola 76th, and Sac 40th. Respectively, Buena Vista, Osceola, and Sac accounted for about 21, four, and 11 percent of HPAI virus-infected commercial poultry farms in IA.

Counties with the greatest number of HPAI virus-infected commercial poultry farms in MN were Kandiyohi, Stearns, and Meeker (Table 6). Kandiyohi is ranked 14th out of 87 counties for the acres of corn grown for grain per county, while Stearns is ranked fifth, and Meeker is ranked 28th. For the square miles of water per county, Kandiyohi is ranked 15th, Stearns 20th, and Meeker 23rd. For the number of turkey farms per county, Kandiyohi is ranked fourth, Stearns first, and Meeker 15th. For the number of laying hen farms per county, Kandiyohi is ranked 35th, Stearns first, and Meeker 14th. Finally, for the number of pullet farms per county, Kandiyohi is ranked 30th, Stearns 15th, and Meeker 27th. Respectively, Kandiyohi, Stearns, and Meeker accounted for about 37, 13, and nine percent of all HPAI virus-infected commercial poultry farms in MN.

Counties with the greatest number of HPAI virus-infected commercial poultry farms in IA were Buena Vista, Sioux, and Sac. Buena Vista is ranked 28th out of 99 counties for the acres of corn grown for grain per county, while Sioux is ranked 6th, and Sac is ranked 25th. For the square miles of water per county, Buena Vista is ranked 11th, Sioux 84th, and Sac 81st. For the number of turkey farms per county, Buena Vista is ranked first, Sioux 69th, and Sac second. For the number of laying hen farms per county, Buena Vista is ranked 97th, Sioux eighth, and Sac 88th. Finally, for the number of pullet farms per county, Buena Vista is ranked 53rd, Sioux third, and Sac 40th. Respectively, Buena Vista, Sioux, and Sac accounted for about 21, 20, and 11 percent of all HPAI virus-infected commercial poultry farms in IA.

CHAPTER 4: DISCUSSION

This study summarized the epidemiologic features of the outbreaks in MN and IA and, using a case-control logistic regression analysis, addressed factors which may have potentially contributed to the outbreak. In MN, it was observed that one county had about a three-fold difference in the number of infected farms relative to other counties. In IA, two counties had about a two-fold difference in the number of infected farms compared to other counties. These three counties not only had the greatest number of infected farms but were also infected early and frequently during the observation period. Therefore, analyses with all counties and removing these three counties were performed to determine if these potentially influential observations distorted the regression estimation. The three dichotomous infection status outcomes used involved turkey farms; laying hen and pullet farms combined; and laying hen, pullet, and turkey farms combined. Prior to removing the counties with the greatest number of infected farms, in both the unadjusted and adjusted analyses, associations were found between the percent of the total county area which is corn and the HPAIT outcome for MN (OR: 1.8, $p=0.002$, 95% CI: 1.3-2.7; adjOR: 2.0; $p=0.001$, 95% CI: 1.3-3.1), between the corn variable and HPAIT for IA (OR: 3.2, $p=0.03$, 95% CI: 1.1-9.2; adjOR: 3.3, $p=0.04$, 95% CI: 1.1-10.0; adjOR: 5.1, $p=0.04$, 95% CI: 1.1-23.3; adjOR: 4.1, $p=0.07$, 95% CI: 0.9-18.3), between the corn variable and HPAICOMB for MN (OR: 1.9, $p=0.0007$, 95% CI: 1.3-2.8; adjOR: 2.1, $p=0.0006$, 95% CI: 1.4-3.2), and between the corn variable and HPAICOMB for IA (OR: 1.9, $p=0.02$, 95% CI: 1.1-3.1; adjOR: 1.8, $p=0.02$, 95% CI: 1.1-3.1; adjOR: 1.6, $p=0.08$, 95% CI: 0.9-2.7). For the HPAIC outcome, only univariate analyses were possible. For MN, the percent of total county area which is farm land was significant at $p<0.05$ (OR: 7.8, $p=0.04$, 95% CI: 1.1-56.2), and the percent of total county area which is cropland was significant at $p<0.1$ (OR: 2.6, $p=0.09$, 95% CI: 0.9-7.5). For

IA, the percent of total county area which is cropland (OR: 1.9, $p=0.03$, 95% CI: 1.1-3.3) and the percent of total county area which is farm land (OR: 2.5, $p=0.03$, 95% CI: 1.1-5.5) were both significant at $p<0.05$. It was predicted that the removal of the counties with the highest number of infected farms would attenuate the magnitude and significance of the regression coefficient estimates. After removal, associations were found between the corn variable and HPAIT for MN (OR: 1.8, $p=0.002$, 95% CI: 1.2-2.7; adjOR: 2.0, $p=0.002$, 95% CI: 1.3-3.1), between the corn variable and HPAIT for IA (OR: 3.2, $p=0.04$, 95% CI: 1.1-9.9), between the corn variable and HPAICOMB for MN (OR: 1.9, $p=0.0009$, 95% CI: 1.3-2.8; adjOR: 2.1, $p=0.0007$, 95% CI: 1.4-3.1), and between the corn variable and HPAICOMB for IA (OR: 1.8, $p=0.03$, 95% CI: 1.1-3.1; adjOR: 1.8, $p=0.03$, 95% CI: 1.1-3.1). Again, only univariate analyses were possible for the HPAIC outcome. In the univariate analysis for MN, the percent of total county area which is cropland was significant at $p<0.1$ (OR: 2.5, $p=0.09$, 95% CI: 0.9-7.4), and the percent of total county area which is farm land was significant at $p<0.05$ (OR: 7.7, $p=0.04$, 95% CI: 1.1-56.0). In the univariate analysis for IA, the percent of total county area which is cropland (OR: 1.6, $p=0.08$, 95% CI: 0.9-2.9) and the percent of total county area which is farm land (OR: 2.1, $p=0.07$, 95% CI: 0.9-4.6) were both significant at $p<0.1$. The magnitude and significance of the regression coefficient estimates were generally slightly attenuated by the removal of Kandiyohi County from the MN dataset and of Buena Vista and Sioux Counties from the IA dataset; however, these observations did not notably affect estimates. The percent of the total county area which is corn grown for grain was the most consistently significant exposure across all three outcome variables. The percent of the total county area which is corn grown for grain was the most consistently significant exposure across all three outcome variables.

4.1 Temporal Distribution

Generally, farms in central MN became infected first, followed by farms in southern MN, northern IA, and central IA (Figure 10). Few farms in southern IA became infected. Most corn appears to be produced in central and southern MN and in northern and central IA (Figures 11, 12). Additionally, domestic poultry production is concentrated in central MN (Figures 13, 14).

Spatially, the counties with the greatest number of HPAI virus-infected commercial poultry farms in MN; Kandiyohi, Stearns, and Meeker; all share borders with one another and accounted for about 59 percent of all HPAI virus-infected commercial poultry farms in MN. For the counties with the greatest number of HPAI virus-infected commercial poultry farms in IA, Buena Vista and Sac share a border, and Buena Vista is one county away from Sioux. These counties accounted for about 52 percent of all HPAI virus-infected commercial poultry farms in IA. Being highly-ranked producers of domestic birds, perhaps the Kandiyohi, Stearns, and Meeker region distributed the virus via supply trucks, equipment or workers moving on and off the farms and through the transport of live birds on major interstate highways to the poultry processing plants. Buena Vista, Sac, and Sioux Counties are also high-ranking domestic bird producers and may have distributed the virus in this manner.

Domestic poultry production and the attraction of migrating wild bird carriers of HPAI virus to areas with high corn production may have impacted the temporal distribution of infected farms. The outbreak appears to be correlated with the major interstate highways leading to the top poultry processing plants and with popular wetland areas for viewing water birds. Several factors related to both poultry production (concentrated poultry production, shipping of live birds to poultry processing plants) and to migratory water birds (corn production, proximity to wet areas) may have played a role in the outbreak. Migratory water birds, which carry the HPAI virus

and are attracted to corn-growing land and water, may have infected domestic poultry in both Canada and the U.S. Movement of equipment to and from regions with concentrated poultry production and the shipping of live birds to poultry processing plants may have contributed to the spread of HPAI.

Results may be generalizable to U.S. counties similar in terms of geography, crop and livestock production, and HPAI susceptibility (e.g. proximity to wild bird migration routes). In terms of HPAI management, if the associations are true, considering these geographic and agricultural factors as potential predictors of HPAI virus spread during an outbreak may be the only practical use of these results. Any modification of agricultural land or wetland to control HPAI is not a practical option. In particular, modification of wetland may cause infected birds to scatter⁶⁴. Wetland also lessens flood damage, filters water, and serves as a habitat for several species⁶⁹. The Ramsar Handbook for the Wise Use of Wetlands recommends consulting zoonoses epidemiology, wildlife, ornithological, and wetland specialists when combating HPAI virus infection⁶⁴. The environment is one aspect considered when determining the extent of an infection control region⁸³. Perhaps HPAI virus infection responders should give more consideration to an environmental factor outside of establishing an infection control area. Accounting for this factor outside of establishing an infection control area may contribute to controlling the outbreak quickly and mitigating its impact.

The transmission ability of current HPAI strains among humans is still not a concern in the U.S., but outbreaks among domestic poultry should be well documented and analyzed in order to minimize the opportunity for strain mixing and mutation, especially in agricultural areas producing large numbers of different domestic animal species. IA and MN were ranked first and third, respectively, in the 2015 ranking of states by swine inventory (Figure 15)⁷⁷. Pigs are

susceptible to multiple influenza strains, including avian and human influenza^{31,81}. Different strains can reassort and give rise to strains which are more transmissible to and among humans and other animals^{4,31,60}. The CDC MMWR has documented outbreaks of both swine and avian influenza viruses in Minnesota and Iowa over the years and described the transmission ability of the 2009 pandemic swine influenza virus among humans. The potential for the mutation and reassortment of influenza and the noted resistance of influenza to antiviral drugs continues to be alarming⁶⁶.

4.2 Strengths

The USDA has examined the U.S. as a whole with regard to the outbreak. It has conducted surveys of domestic poultry operation staff and investigated factors such as wind which may have contributed to the outbreak; however, it has not specifically focused on agricultural and geographic factors, which have a role in HPAI virus transmission. This study evaluated the possible association between geographic and agricultural traits of counties in the two most affected states and county infection status. All exposure variables were continuous in the analysis.

4.3 Future Research

Future studies should examine the environmental portion of the HPAI virus transmission pathway more closely and clarify the size of the role of geographic and agricultural factors in transmission. Studies should be conducted on the farm level so that more specific inferences can be made. With more complete data, an outcome involving the proportion of farms in a county that are infected should be used in linear regression.

APPENDICES

APPENDIX A:

Tables

Table 1a: Distribution of infected farms among states and individual MN and IA counties in the USDA APHIS “HPAI 2014-2015 Infected Premises” (state, county, number of birds, flock type, species, HPAI strain confirmed) dataset (N=232 infected farms) and the analytic sample (N=180 infected farms)

	USDA APHIS Dataset (Max N=232 infected farms)		Infected Farms Analytic Sample (Max N=180 infected farms)	
	N	(%)	N	(%)
State				
Arkansas	1	0.43	0	0
California	2	0.86	0	0
Iowa	77	33.19	71	39.44
Idaho	1	0.43	0	0
Indiana	1	0.43	0	0
Kansas	1	0.43	0	0
Minnesota	110	47.41	109	60.56
Missouri	3	1.29	0	0
Montana	1	0.43	0	0
North Dakota	2	0.86	0	0
Nebraska	6	2.59	0	0
Oregon	2	0.86	0	0
South Dakota	10	4.31	0	0
Washington	5	2.16	0	0
Wisconsin	10	4.31	0	0

Table 1a (cont'd)

	USDA APHIS Dataset Iowa and Minnesota (Max N=187 infected farms)		Infected Farms Analytic Sample Excluding Backyard Farms (Max N=180 infected farms)	
	N	(%)	N	(%)
County				
<u>Iowa:</u>				
Adair	1	1.30	1	1.41
Buena Vista	15	19.48	15	21.13
Calhoun	2	2.60	2	2.82
Cherokee	5	6.49	5	7.04
Clay	2	2.60	2	2.82
Hamilton	4	5.19	4	5.63
Kossuth	1	1.30	1	1.41
Lyon	1	1.30	1	1.41
Madison	1	1.30	1	1.41
O'Brien	3	3.90	2	2.82
Osceola	4	5.19	3	4.22
Palo Alto	1	1.30	1	1.41
Plymouth	1	1.30	1	1.41
Pocahontas	3	3.90	3	4.22
Sac	8	10.39	8	11.27
Sioux	18	23.38	14	19.72
Webster	1	1.30	1	1.41
Wright	6	7.79	6	8.45

Table 1a (cont'd)

	USDA APHIS Dataset Iowa and Minnesota (Max N=187 infected farms)		Infected Farms Analytic Sample Excluding Backyard Farms (Max N=180 infected farms)	
	N	(%)	N	(%)
County				
<u>Minnesota:</u>				
Blue Earth	1	0.91	1	0.92
Brown	5	4.55	5	4.59
Chippewa	1	0.91	1	0.92
Clay	1	0.91	1	0.92
Cottonwood	2	1.82	2	1.83
Kandiyohi	40	36.36	40	36.70
Lac Qui Parle	1	0.91	1	0.92
Le Sueur	1	0.91	1	0.92
Lyon	1	0.91	1	0.92
Meeker	10	9.09	10	9.17
Nicollet	1	0.91	1	0.92
Nobles	1	0.91	1	0.92
Otter Tail	4	3.64	4	3.67
Pipestone	2	1.82	1	0.92
Pope	2	1.82	2	1.83
Redwood	4	3.64	4	3.67
Renville	8	7.27	8	7.34
Roseau	1	0.91	1	0.92
Stearns	14	12.73	14	12.84
Steele	1	0.91	1	0.92
Swift	7	6.36	7	6.42
Wadena	1	0.91	1	0.92
Watonwan	1	0.91	1	0.92

Table 1b: Characteristics of infected farms in MN and IA counties in the USDA APHIS “HPAI 2014-2015 Infected Premises” (state, county, number of birds, flock type, species, HPAI strain confirmed) dataset (N=232 infected farms) and the analytic sample (N=180 infected farms)

	USDA APHIS Dataset (Max N=232 infected farms)		Infected Farms Analytic Sample Excluding Backyard Farms (Max N=180 infected farms)	
	N	(%)	N	(%)
Flock Type				
commercial	180	96.26	180	100
backyard	7	3.74	0	0
Number of Birds				
0-100,000	142	76.76	135	75.84
>100,000-200,000	11	5.95	11	6.18
>200,000-300,000	10	5.41	10	5.62
>300,000-400,000	4	2.16	4	2.25
>400,000-500,000	2	1.08	2	1.12
>500,000	16	8.65	16	8.99
Species				
laying hen	27	14.44	27	15
turkey	139	74.33	139	77.22
pullet	14	7.49	14	7.78
other	7	3.74	0	0
HPAI Strain				
H5N2	179	95.72	173	96.11
H5	2	1.07	1	0.56
DCHP	6	3.21	6	3.33

USDA=United States Department of Agriculture

APHIS=Animal and Plant Health Inspection Service

HPAI=highly pathogenic avian influenza

DCHP=Dangerous Contact Highly Pathogenic

Table 2: Characteristics of Minnesota and Iowa counties in the 2010 U.S. Census[†] and the 2012 USDA Census of Agriculture^{*} datasets

	N of Counties	TOTAREAC [†] Mean/Median (Range)	WATAC [†] Mean/Median (Range)	FARMAC [*] Mean/Median (Range)	CROPAC [*] Mean/Median (Range)	CORNGRAINAC [*] Mean/Median (Range)	ALLPOUFA [*] Mean/Median (Range)	LAYFA [*] Mean/Median (Range)	TURKFA [*] Mean/Median (Range)	PULFA [*] Mean/Median (Range)
MN	87	639552/45894 4 (4281216) <u>min</u> 108864 <u>max</u> 4390080 m=0	53760/11008 (1207808) <u>min</u> 57.6 <u>max</u> 1207872 m=0	299262.5/271737 (1094233) <u>min</u> 723 <u>max</u> 1094956 m=0	248242.9/225497 (991220) <u>min</u> 185 <u>max</u> 991405 m=0	96334.6/94040 (284965) <u>min</u> 0 <u>max</u> 284965 m=2	62.1/45 (360) <u>min</u> 1 <u>max</u> 361 m=0	51.7/39 (263) <u>min</u> 0 <u>max</u> 263 m=0	6.4/5 (38) <u>min</u> 0 <u>max</u> 38 m=0	6.2/5 (36) <u>min</u> 0 <u>max</u> 36 m=0
IA	99	363776/36651 5.2 (367955.2) <u>min</u> 255609.6 <u>max</u> 623564.8 m=0	2687/1324.8 (14924.8) <u>min</u> 25.6 <u>max</u> 14950.4 m=0	309321/312457 (430899) <u>min</u> 168540 <u>max</u> 599439 m=0	265216/273269 (469685) <u>min</u> 98944 <u>max</u> 568629 m=0	138478.9/1413 11 (301626) <u>min</u> 22854 <u>max</u> 324480 m=0	43.8/38 (113) <u>min</u> 11 <u>max</u> 124 m=0	38.6/32 (105) <u>min</u> 10 <u>max</u> 115 m=0	4.1/3 (18) <u>min</u> 0 <u>max</u> 18 m=0	5/4 (32) <u>min</u> 0 <u>max</u> 32 m=0

Missing Data (2012 USDA Census of Agriculture dataset): corn grown for grain per county (MN, N=2)

USDA=United States Department of Agriculture

m=missing

MN=Minnesota

IA=Iowa

INF=infected

UNINF=uninfected

TOTAREAC=acres of total area per county

WATAC=acres of water per county

FARMAC=acres of farm land per county

CROPAC=acres of total cropland per county

CORNGRAINAC=acres of corn grown for grain per county

ALLPOUFA=number of all types of poultry farms per county

LAYFA=number of laying hen farms per county

TURKFA=number of turkey farms per county

PULFA=number of pullet farms per county

Table 2 (cont'd)

	N of Counties	TOTAREAC [†] Mean/Median (Range)	WATAC [†] Mean/Median (Range)	FARMAC [†] Mean/Median (Range)	CROPAC [†] Mean/Median (Range)	CORNGRAINAC [†] Mean/Median (Range)	ALLPOUFA [†] Mean/Median (Range)	LAYFA [†] Mean/Median (Range)	TURKFA [†] Mean/Median (Range)	PULFA [†] Mean/Median (Range)
UN- INF MN	64	680809/45626 9 (4281229) <u>min</u> 108851 <u>max</u> 4390080 m=0	67010.8/13452 .8 (1207795) <u>min</u> 57.6 <u>max</u> 1207853 m=0	259999/234128 (1094233) <u>min</u> 723 <u>max</u> 1094956 m=0	210853/194995 (991220) <u>min</u> 185 <u>max</u> 991405 m=0	75530.4/62315 (237118) <u>min</u> 0 <u>max</u> 237118 m=2	62.1/50.5 (258) <u>min</u> 1 <u>max</u> 259 m=0	53.3/44.5 (164) <u>min</u> 0 <u>max</u> 164 m=0	5.5/4 (35) <u>min</u> 0 <u>max</u> 35 m=0	6.4/5 (36) <u>min</u> 0 <u>max</u> 36 m=0
INF MN	23	639528/45894 4 (4281229) <u>min</u> 108851 <u>max</u> 4390080 m=0	53768.4/11027 .2 (1207795) <u>min</u> 57.6 <u>max</u> 1207853 m=0	299262.5/271737 (1094233) <u>min</u> 723 <u>max</u> 1094956 m=0	248242.9/225497 (991220) <u>min</u> 185 <u>max</u> 991405 m=0	96334.6/94040 (284965) <u>min</u> 0 <u>max</u> 284965 m=0	62.1/45 (360) <u>min</u> 1 <u>max</u> 361 m=0	51.7/39 (263) <u>min</u> 0 <u>max</u> 263 m=0	6.4/5 (38) <u>min</u> 0 <u>max</u> 38 m=0	6.2/5 (36) <u>min</u> 0 <u>max</u> 36 m=0
UN- INF IA	81	356080/36512 6 (356742) <u>min</u> 257101 <u>max</u> 613843 m=0	3008.1/1376 (14924.8) <u>min</u> 25.6 <u>max</u> 14950.4 m=0	295877.4/299179 (364293) <u>min</u> 168540 <u>max</u> 532833 m=0	249596.9/262152 (382885) <u>min</u> 98944 <u>max</u> 481829 m=0	129695.4/1322 62 (232467) <u>min</u> 22854 <u>max</u> 255321 m=0	45.3/40 (113) <u>min</u> 11 <u>max</u> 124 m=0	40.6/36 (104) <u>min</u> 11 <u>max</u> 115 m=0	3.9/3 (15) <u>min</u> 0 <u>max</u> 15 m=0	5.2/4 (32) <u>min</u> 0 <u>max</u> 32 m=0
INF IA	18	363784/36651 5 (367955) <u>min</u> 255610 <u>max</u> 623565 m=0	2687.1/1324.8 (14924.8) <u>min</u> 25.6 <u>max</u> 14950.4 m=0	309320.5/312457 (430899) <u>min</u> 168540 <u>max</u> 599439 m=0	265215.6/273269 (469685) <u>min</u> 98944 <u>max</u> 568629 m=0	138478.9/1413 11 (301626) <u>min</u> 22854 <u>max</u> 324480 m=0	43.8/38 (113) <u>min</u> 11 <u>max</u> 124 m=0	38.6/32 (105) <u>min</u> 10 <u>max</u> 115 m=0	4.1/3 (18) <u>min</u> 0 <u>max</u> 18 m=0	5/4 (32) <u>min</u> 0 <u>max</u> 32 m=0

Missing Data (2012 USDA Census of Agriculture dataset): corn grown for grain per county (MN, N=2)

USDA=United States Department of Agriculture

m=missing

MN=Minnesota

IA=Iowa

INF=infected

UNINF=uninfected

TOTAREAC=acres of total area per county

WATAC=acres of water per county

FARMAC=acres of farm land per county

CROPAC=acres of total cropland per county

CORNGRAINAC=acres of corn grown for grain per county

ALLPOUFA=number of all types of poultry farms per county

LAYFA=number of laying hen farms per county

TURKFA=number of turkey farms per county

PULFA=number of pullet farms per county

Table 3: Explanation of acronyms

USDA	United States Department of Agriculture
APHIS	Animal and Plant Health Inspection Service
HPAI	highly pathogenic avian influenza
LPAI	low pathogenic avian influenza
HPAIT	highly pathogenic avian influenza infection status, where 0=no turkey farms with virus infection and 1=at least one infected turkey farm
HPAIC	highly pathogenic avian influenza infection status, where 0=no laying hen or pullet farms with virus infection and 1=at least one infected laying hen or pullet farm
HPAICOMB	highly pathogenic avian influenza infection status, where 0=no laying hen or pullet or turkey farms with virus infection and 1=at least one infected laying hen or pullet or turkey farm
ALLPOUFA	number of all types of poultry farms per county
TURKFA	number of turkey farms per county
LAYFA	number of laying hen farms per county
PULFA	number of pullet farms per county
LAYPULFA	combined number of laying hen and pullet farms per county
TOTAREAC	acres of total area per county
CROPAC	acres of total cropland per county
CROPPRC	percent of total county area which is cropland
CORNGRAINAC	acres of corn grown for grain per county
CORNGRPRC	percent of total county area which is corn grown for grain
FARMAC	acres of farm land per county
FARMPRC	percent of total county area which is farm land
WATAC	acres of water per county
WATSQPRC	percent of total county area which is water

Table 4: Description of the highly pathogenic avian influenza outbreak in Minnesota and Iowa between April and June of 2015

State	Species	At-risk at Beginning of Period	Developed HPAI H5N2 Virus Infection by End of Period	Cumulative Incidence (per 100 over 91 Days)	Incidence Rate (per 100,000 Flock-days of Observation) [95% CI ^d]
IA	laying hen	3821 flocks 52,218,870 birds	22 flocks 25,892,745 birds	22/3821 = 0.576	22/346552 flock-days = 6.348 [3.977, 9.612]
	turkey	402 flocks 4,383,172 birds	35 flocks 1,146,586 birds	35/402 = 8.706	35/34878 flock-days = 100.350 [69.89, 139.6]
	pullet	499 flocks 12,565,630 birds	13 flocks 5,609,496 birds	13/499 = 2.605	13/44778 flock-days = 29.030 [15.44, 49.65]
MN	laying hen	4501 flocks 9,693,648 birds	4 flocks 3,625,677 birds	4/4501 = 0.0889	4/409355 flock-days = 0.977 [0.263, 2.502]
	turkey	559 flocks 19,449,992 birds	94 flocks 4,664,072 birds	94/559 = 16.816	94/44937 flock-days = 209.182 [169, 256]
	pullet	539 flocks 2,823,994 birds	1 flock 410,514 birds	1/539 = 0.186	1/49021 flock-days = 2.0399 [0.02668, 11.35]
^d Byar's approximation of exact Poisson confidence interval					

Species	Incidence Rate Ratio (MN/IA)	95% CI ^e
laying hen	0.154	0.053-0.45 [†]
turkey	2.085	1.41-3.07 [†]
pullet	0.0703	0.0092-0.54 [†]
^e Byar's approximation of exact Poisson confidence interval		
[†] p < 0.0001; [†] p < 0.001		

HPAI=highly pathogenic avian influenza

IA=Iowa

MN=Minnesota

Table 5a: Results of logistic regression with HPAIC outcome based on data from all counties

Outcome (HPAIC): 0 = no laying hen or pullet farms with HPAI virus infection 1 = at least one infected laying hen or pullet farm							
Model	Variable	ML Estimate	ML SE	Wald χ^2	Pr> χ^2	OR	CL
MN							
<i>uni-variable</i>							
	TURKFA	0.065	0.050	1.67	0.20	1.07	0.97, 1.18
	LAYPULFA	0.011	0.0077	2.02	0.16	1.01	0.99, 1.03
	CROPPRC	0.094	0.055	2.88	0.090 [†]	2.55 ^a	0.87, 7.50
	CORNGRPC	0.070	0.044	2.52	0.11	2.01 ^a	0.85, 4.78
	FARMPRC	0.21	0.10	4.15	0.042 [*]	7.79 ^a	1.08, 56.17
	WATSQPRC	-0.20	0.22	0.78	0.38	0.14 ^a	0.0020, 11.07
<i>multi-variable</i>							
	N/A						
IA							
<i>uni-variable</i>							
	TURKFA	-0.019	0.086	0.050	0.82	0.98	0.83, 1.16
	LAYPULFA	-0.011	0.014	0.53	0.47	0.99	0.96, 1.02
	CROPPRC	0.062	0.029	4.53	0.033 [*]	1.86 ^a	1.05, 3.28
	CORNGRPC	0.035	0.027	1.68	0.19	1.42 ^a	0.84, 2.43
	FARMPRC	0.091	0.041	5.04	0.025 [*]	2.49 ^a	1.12, 5.51
	WATSQPRC	-1.59	1.02	2.43	0.12	<0.0010 ^a	<0.0010, 59.54
<i>multi-variable</i>							
	N/A						

*N/A indicates that no variables remained statistically significant after mutual adjustment in the multivariate model

Table 5a (cont'd)

HPAI=highly pathogenic avian influenza

HPAIC=HPAI virus infection status, where 0=no laying hen or pullet farms with HPAI virus infection and 1=at least one infected laying hen or pullet farm

HPAIT=HPAI virus infection status, where 0=no turkey farms with HPAI virus infection and 1=at least one infected turkey farm

HPAICOMB=highly pathogenic avian influenza infection status, where 0=no laying hen or pullet or turkey farms with virus infection and 1=at least one infected laying hen or pullet or turkey farm

MN=Minnesota

IA=Iowa

ML=maximum likelihood

SE=standard error

X²=chi-square

OR=odds ratio

CL=95% Wald confidence limits

^ap<0.001; ^bp<0.01; ^cp<0.05; ^dp<0.1; ^eper 10 units

TURKFA=number of turkey farms per county

LAYFA=number of laying hen farms per county

PULFA=number of pullet farms per county

LAYPULFA=combined number of laying hen and pullet farms per county

CROPPRC=percent of total county area which is cropland

CORNGRPC=percent of total county area which is corn grown for grain

FARMPRC=percent of total county area which is farm land

WATSQPRC=percent of total county area which is water

Table 5aa: Results of logistic regression with HPAIC outcome following removal of Kandiyohi (MN), Buena Vista (IA), and Sioux (IA) Counties

Outcome (HPAIC): 0 = no laying hen or pullet farms with HPAI virus infection 1 = at least one infected laying hen or pullet farm							
Model	Variable	ML Estimate	ML SE	Wald X ²	Pr>X ²	OR	CL
MN							
<i>uni-variable</i>							
	TURKFA	0.068	0.050	1.86	0.17	1.07	0.97, 1.18
	LAYPULFA	0.011	0.0077	2.00	0.16	1.01	0.996, 1.03
	CROPPRC	0.092	0.055	2.82	0.093 [†]	2.52 ^a	0.86, 7.39
	CORNGRPC	0.070	0.044	2.53	0.11	2.01 ^a	0.85, 4.72
	FARMPRC	0.20	0.10	4.07	0.044 [*]	7.70 ^a	1.06, 55.96
	WATSQPRC	-0.19	0.22	0.74	0.39	0.15 ^a	0.002, 11.52
<i>multi-variable</i>							
	N/A						
IA							
<i>uni-variable</i>							
	TURKFA	-0.17	0.13	1.57	0.21	0.85	0.65, 1.10
	LAYPULFA	-0.018	0.018	1.06	0.30	0.98	0.95, 1.02
	CROPPRC	0.050	0.029	2.98	0.084 [†]	1.64 ^a	0.94, 2.88
	CORNGRPC	0.029	0.028	1.03	0.31	1.34 ^a	0.76, 2.33
	FARMPRC	0.073	0.040	3.28	0.070 [†]	2.08 ^a	0.94, 4.57
	WATSQPRC	-2.22	1.42	2.45	0.12	<0.0010 ^a	<0.0010, 268.50
<i>multi-variable</i>							
	N/A						

*N/A indicates that no variables remained statistically significant after mutual adjustment in the multivariate model

Table 5b: Results of logistic regression with HPAIT outcome based on data from all counties

Outcome (HPAIT): 0 = no turkey farms with HPAI virus infection 1 = at least one infected turkey farm							
Model	Variable	ML Estimate	ML SE	Wald X ²	Pr>X ²	OR	CL
MN							
<i>uni-variable</i>							
	TURKFA	0.087	0.038	5.13	0.024*	1.09	1.01, 1.18
	LAYPULFA	-0.0021	0.0055	0.14	0.71	0.99	0.99, 1.01
	CROPPRC	0.036	0.012	8.30	0.0040 [◇]	1.43 ^a	1.12, 1.82
	CORNGRPC	0.060	0.019	9.67	0.0019 [◇]	1.83 ^a	1.25, 2.67
	FARMPRC	0.042	0.015	7.68	0.0056 [◇]	1.53 ^a	1.13, 2.06
	WATSQPRC	-0.14	0.082	2.80	0.094 [†]	0.25 ^a	0.051, 1.26
<i>multi-variable</i>							
	TURKFA	0.10	0.041	6.24	0.013*	1.11	1.02, 1.20
	CORNGRPC	0.070	0.022	10.20	0.0014 [◇]	2.02 ^a	1.31, 3.10

Table 5b (cont'd)

Outcome (HPAIT): 0 = no turkey farms with HPAI virus infection 1 = at least one infected turkey farm							
Model	Variable	ML Estimate	ML SE	Wald χ^2	Pr> χ^2	OR	CL
IA							
<i>uni-variable</i>							
	TURKFA	0.25	0.086	8.29	0.0040 [◇]	1.28	1.08, 1.52
	LAYPULFA	-0.11	0.047	5.06	0.025 [*]	0.90	0.82, 0.99
	CROPPRC	0.15	0.069	4.88	0.027 [*]	4.55 ^a	1.19, 17.43
	CORNGRPC	0.12	0.054	4.74	0.029 [*]	3.21 ^a	1.12, 9.17
	FARMPRC	0.14	0.068	4.42	0.035 [*]	4.16 ^a	1.10, 15.71
	WATSQPRC	-0.51	0.65	0.61	0.43	0.0060 ^a	<0.0010, >999.99
<i>multi-variable</i>							
	TURKFA	0.36	0.16	5.22	0.022 [*]	1.43	1.05, 1.94
	LAYPULFA	-0.15	0.070	4.85	0.028 [*]	0.86	0.75, 0.98
	CORNGRPC	0.14	0.077	3.30	0.069 [†]	4.05 ^a	0.90, 18.34
	TURKFA	0.23	0.091	6.69	0.0097 [◇]	1.26	1.06, 1.51
	CORNGRPC	0.12	0.057	4.30	0.038 [*]	3.27 ^a	1.07, 9.99
	LAYPULFA	-0.14	0.065	4.72	0.030 [*]	0.87	0.77, 0.99
	CORNGRPC	0.16	0.077	4.45	0.035 [*]	5.11 ^a	1.12, 23.29

Table 5bb: Results of logistic regression with HPAIT outcome following removal of Kandiyohi (MN), Buena Vista (IA), and Sioux (IA) Counties

Outcome (HPAIT): 0 = no turkey farms with HPAI virus infection 1 = at least one infected turkey farm							
Model	Variable	ML Estimate	ML SE	Wald X ²	Pr>X ²	OR	CL
MN							
<i>uni- variable</i>							
	TURKFA	0.076	0.038	4.00	0.046*	1.08	1.00, 1.16
	LAYPULFA	-0.0021	0.0056	0.15	0.70	0.99	0.99, 1.01
	CROPPRC	0.036	0.013	8.07	0.0045 [◇]	1.43 ^a	1.12, 1.83
	CORNGRPRC	0.060	0.020	9.31	0.0023 [◇]	1.82 ^a	1.24, 2.67
	FARMPRC	0.043	0.016	7.44	0.0064 [◇]	1.53 ^a	1.13, 2.08
	WATSQPRC	-0.17	0.094	3.25	0.072 [†]	0.18 ^a	0.029, 1.16
<i>multi- variable</i>							
	TURKFA	0.094	0.041	5.15	0.023*	1.10	1.01, 1.19
	CORNGRPRC	0.069	0.022	9.90	0.0017 [◇]	1.99 ^a	1.30, 3.06

Table 5bb (cont'd)

Outcome (HPAIT): 0 = no turkey farms with HPAI virus infection 1 = at least one infected turkey farm							
Model	Variable	ML Estimate	ML SE	Wald χ^2	Pr> χ^2	OR	CL
IA							
<i>uni-variable</i>							
	TURKFA	0.21	0.096	4.55	0.033*	1.23	1.02, 1.48
	LAYPULFA	-0.089	0.045	3.83	0.0502 [†]	0.92	0.84, 1.00
	CROPPRC	0.14	0.069	4.27	0.039*	4.15 ^a	1.08, 15.97
	CORNGRPC	0.12	0.057	4.22	0.040*	3.23 ^a	1.06, 9.91
	FARMPRC	0.13	0.069	3.72	0.054 [†]	3.77 ^a	0.98, 14.53
	WATSQPRC	-0.78	0.89	0.78	0.38	<0.0010 ^a	<0.0010, >999.99
<i>multi-variable</i>							
	N/A						

*N/A indicates that no variables remained statistically significant after mutual adjustment in the multivariate model

Table 5c: Results of logistic regression with HPAICOMB outcome based on data from all counties

Outcome (HPAICOMB): 0 = no laying hen or pullet or turkey farms with HPAI virus infection 1 = at least one infected laying hen or pullet or turkey farm							
Model	Variable	ML Estimate	ML SE	Wald χ^2	Pr> χ^2	OR	CL
MN							
<i>uni-variable</i>							
	TURKFA	0.070	0.036	3.76	0.052 [†]	1.07	0.99, 1.15
	LAYPULFA	-0.0032	0.0054	0.34	0.56	0.99	0.99, 1.01
	CROPPRC	0.041	0.013	10.26	0.0014 [◊]	1.51 ^a	1.17, 1.94
	CORNGRPC	0.066	0.019	11.47	0.00070 ^Δ	1.93 ^a	1.32, 2.82
	FARMPRC	0.050	0.016	9.14	0.0025 [◊]	1.64 ^a	1.19, 2.27
	WATSQPRC	-0.15	0.081	3.24	0.072 [†]	0.24 ^a	0.048, 1.14
<i>multi-variable</i>							
	TURKFA	0.084	0.039	4.75	0.029 [*]	1.09	1.01, 1.17
	CORNGRPC	0.073	0.021	11.82	0.00060 ^Δ	2.08 ^a	1.37, 3.15

Table 5c (cont'd)

Outcome (HPAICOMB): 0 = no laying hen or pullet or turkey farms with HPAI virus infection 1 = at least one infected laying hen or pullet or turkey farm							
Model	Variable	ML Estimate	ML SE	Wald X^2	Pr> X^2	OR	CL
IA							
<i>uni-variable</i>							
	TURKFA	0.070	0.064	1.19	0.27	1.07	0.95, 1.22
	LAYPULFA	-0.027	0.015	3.22	0.073 [†]	0.97	0.95, 1.00
	CROPPRC	0.088	0.029	9.17	0.0025 [◇]	2.42 ^a	1.37, 4.28
	CORNGRPC	0.062	0.026	5.66	0.017 [*]	1.87 ^a	1.12, 3.12
	FARMPRC	0.11	0.037	9.14	0.0025 [◇]	3.09 ^a	1.49, 6.43
	WATSQPRC	-1.32	0.73	3.30	0.069 [†]	<0.0010 ^a	<0.0010, 2.84
<i>multi-variable</i>							
	LAYPULFA	-0.026	0.015	2.97	0.085 [†]	0.97	0.95, 1.00
	CORNGRPC	0.061	0.027	5.31	0.021 [*]	1.84 ^a	1.10, 3.10

Table 5cc: Results of logistic regression with HPAICOMB outcome following removal of Kandiyohi (MN), Buena Vista (IA), and Sioux (IA) Counties

Outcome (HPAICOMB): 0 = no laying hen or pullet or turkey farms with HPAI virus infection 1 = at least one infected laying hen or pullet or turkey farm							
Model	Variable	ML Estimate	ML SE	Wald χ^2	Pr> χ^2	OR	CL
MN							
<i>uni-variable</i>							
	TURKFA	0.060	0.036	2.74	0.098 [†]	1.06	0.99, 1.14
	LAYPULFA	-0.0033	0.0055	0.35	0.55	0.997	0.99, 1.01
	CROPPRC	0.041	0.013	10.02	0.0015 [◇]	1.51 ^a	1.17, 1.95
	CORNGRPC	0.065	0.020	11.10	0.0009 [△]	1.92 ^a	1.31, 2.81
	FARMPRC	0.050	0.017	8.90	0.0029 [◇]	1.65 ^a	1.19, 2.29
	WATSQPRC	-0.17	0.091	3.68	0.055 [†]	0.17 ^a	0.029, 1.04
<i>multi-variable</i>							
	TURKFA	0.076	0.039	3.77	0.052 [†]	1.08	0.999, 1.17
	CORNGRPC	0.072	0.021	11.49	0.00070 [△]	2.05 ^a	1.35, 3.11

Table 5cc (cont'd)

Outcome (HPAICOMB): 0 = no laying hen or pullet or turkey farms with HPAI virus infection 1 = at least one infected laying hen or pullet or turkey farm							
Model	Variable	ML Estimate	ML SE	Wald X²	Pr>X²	OR	CL
IA							
<i>uni-variable</i>							
	TURKFA	0.035	0.075	0.22	0.64	1.04	0.90, 1.20
	LAYPULFA	-0.038	0.018	4.32	0.038*	0.96	0.93, 0.998
	CROPPRC	0.080	0.029	7.69	0.0055 [◇]	2.22 ^a	1.26, 3.90
	CORNGRPC	0.059	0.027	4.86	0.028*	1.81 ^a	1.07, 3.06
	FARMPRC	0.10	0.037	7.44	0.0064 [◇]	2.75 ^a	1.33, 5.69
	WATSQPRC	-1.53	0.86	3.16	0.076 [†]	<0.0010 ^a	<0.0010, 4.80
<i>multi-variable</i>							
	LAYPULFA	-0.038	0.019	4.19	0.041*	0.96	0.93, 0.998
	CORNGRPC	0.059	0.028	4.54	0.033*	1.80 ^a	1.05, 3.10

Table 6: Characteristics of the counties in Minnesota and Iowa with the greatest number of infected farms

COUNTY	TOTARESQMI	CROPAC	CORNGRAINAC	ALLPOUFA	TURKFA	LAYFA	PULFA	FARMAC	WATSQMI
MN									
Kandiyohi	862.35	354055	173617	73	22	52	6	415090	65.57
Stearns	1389.95	582796	229709	361	38	263	11	757637	46.82
Meeker	644.78	259935	122718	100	11	87	7	303795	36.6
IA									
Buena Vista	580.23	333950	176744	36	18	11	3	360849	5.31
Sioux	768.99	442375	219417	86	2	76	16	484491	0.66
Sac	578.33	332957	181714	37	17	18	5	357032	3.32

MIN=Minnesota

IA=Iowa

TOTARESQMI=acres of total area per county

CROPAC=acres of total cropland per county

CORNGRAINAC=acres of corn grown for grain per county

ALLPOUFA=number of all types of poultry farms per county

TURKFA=number of turkey farms per county

LAYFA=number of laying hen farms per county

PULFA=number of pullet farms per county

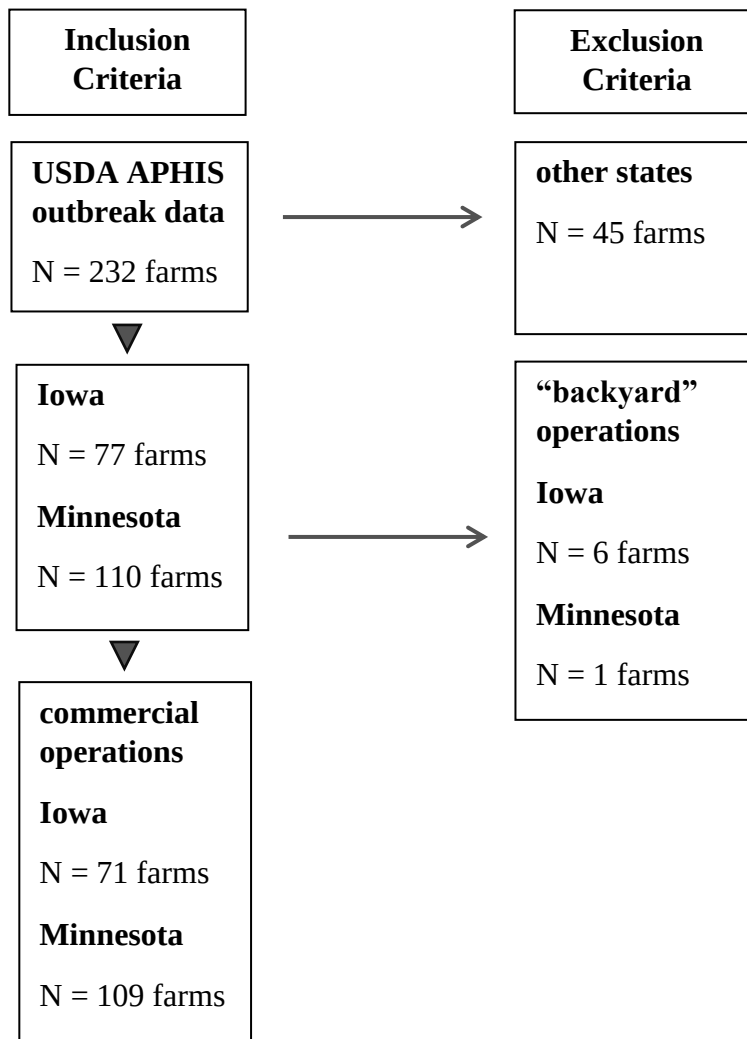
FARMAC=acres of farm land per county

WATSQMI=square miles of water per county

APPENDIX B:

Figures

Figure 1: Derivation of the analytic sample for infected farms



USDA=United States Department of Agriculture

APHIS=Animal and Plant Health Inspection Service

Figure 2: Epidemic curve: Laying hen, turkey, and pullet farms in Minnesota reported as positive for highly pathogenic avian influenza (HPAI) virus during April 2015-June 2015

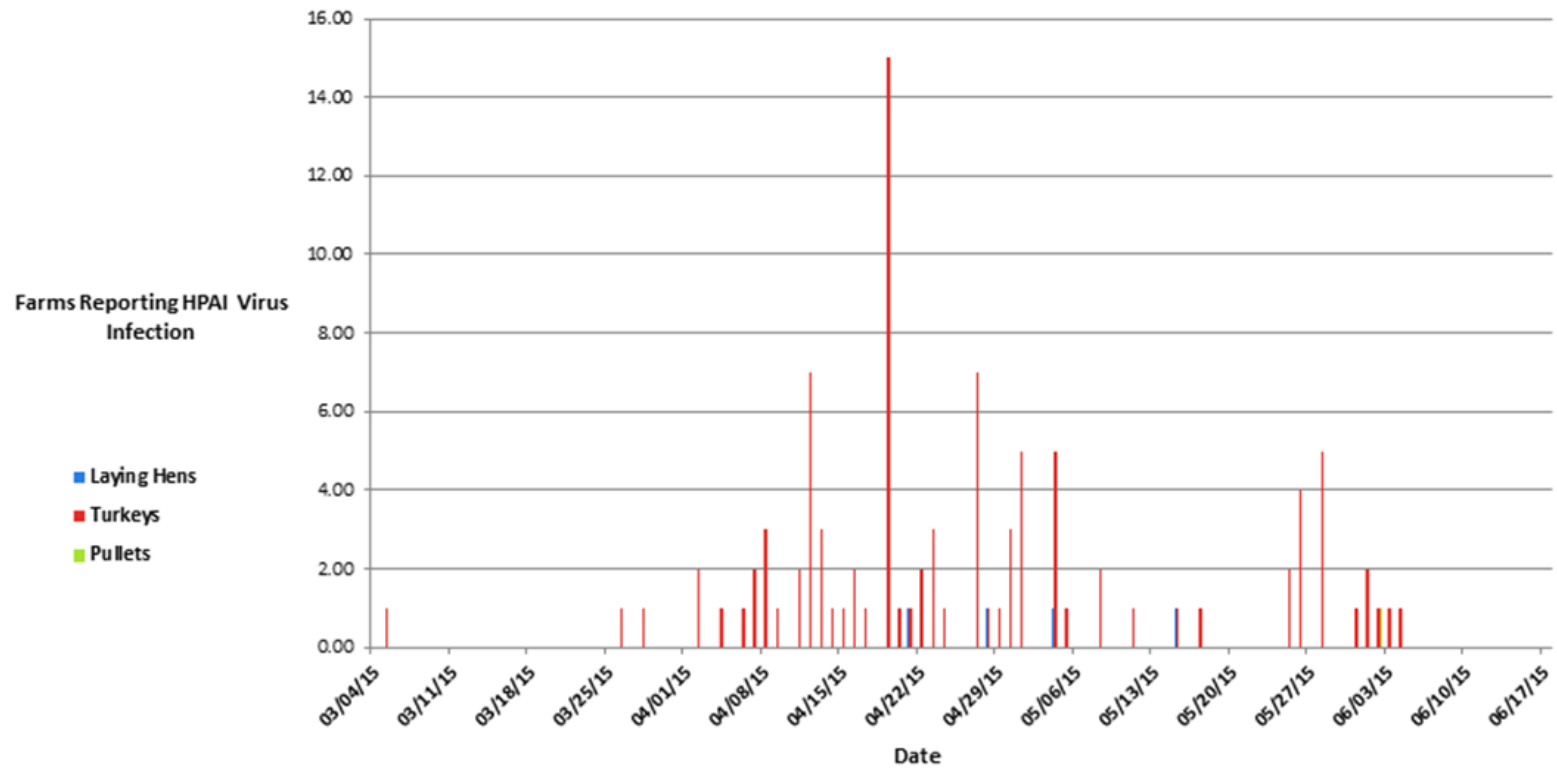


Figure 3: Epidemic curve: Laying hen, turkey, and pullet farms in Iowa reported as positive for highly pathogenic avian influenza (HPAI) virus during April 2015-June 2015

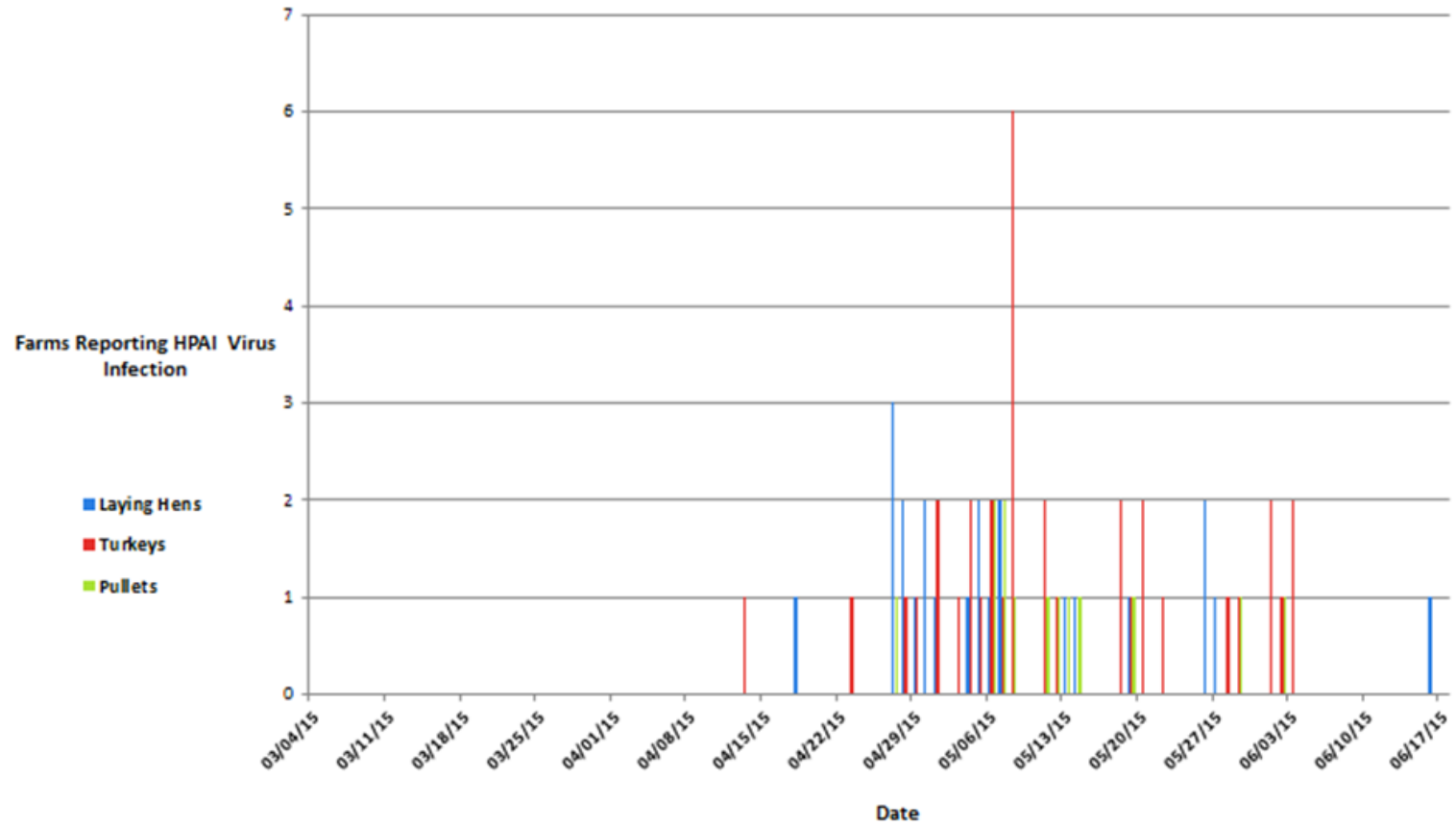
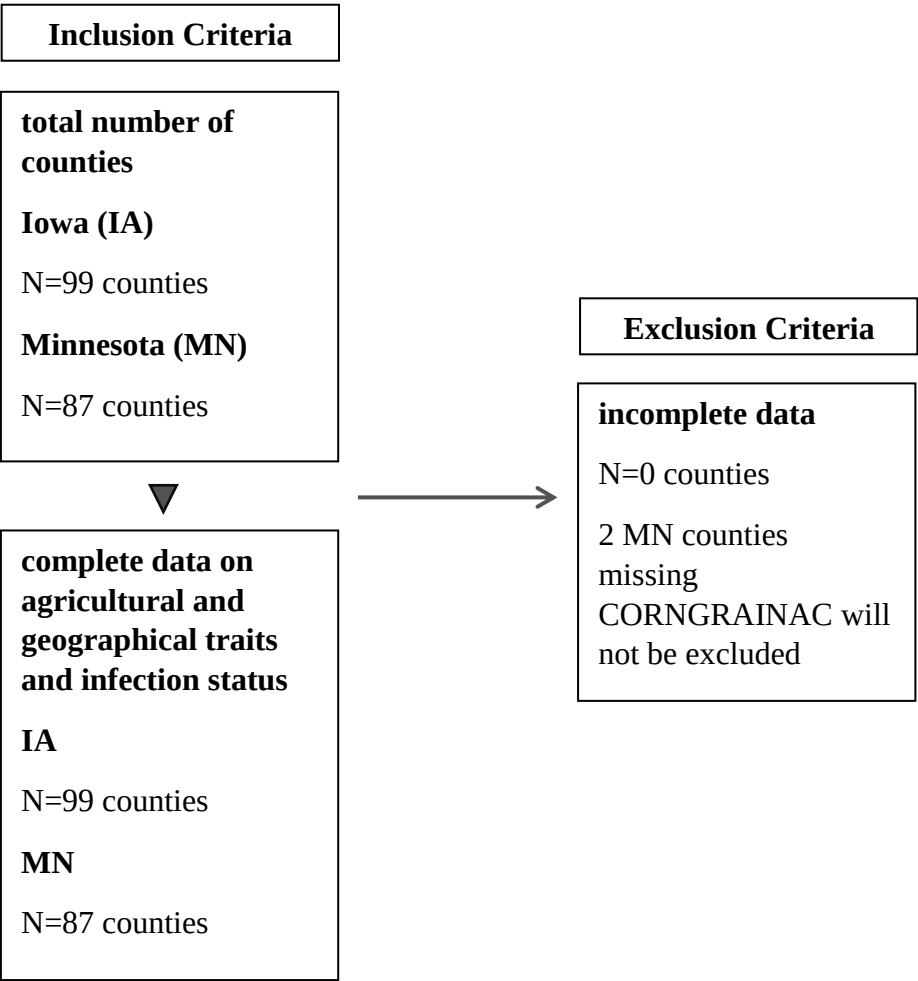


Figure 4: Derivation of the analytic sample for all MN and IA counties



IA=Iowa

MN=Minnesota

CORNGRAINAC=acres of corn grown for grain per county

Figure 5: Minnesota counties with at least one infected turkey, laying hen, or pullet farm

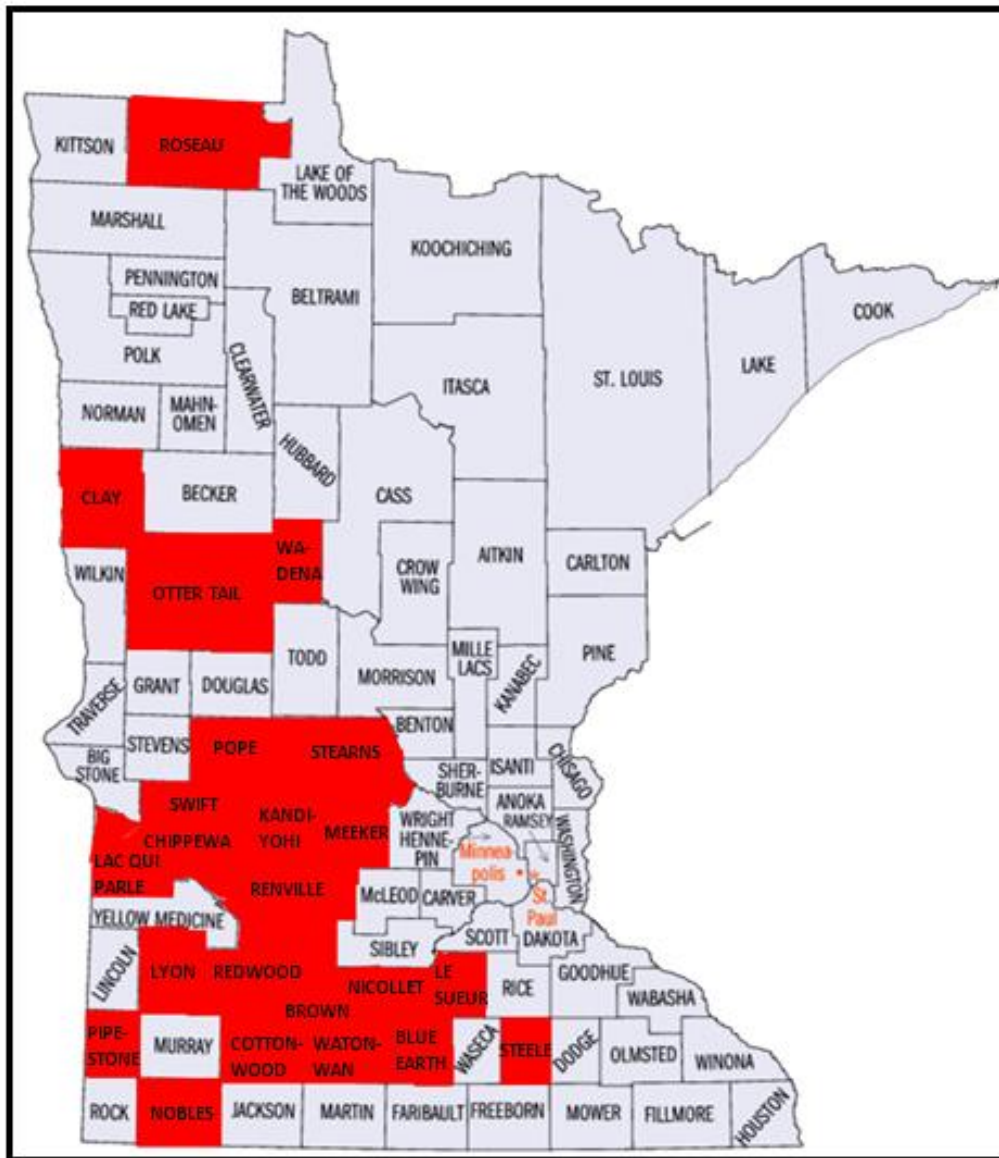


Figure 6: Iowa counties with at least one infected turkey, laying hen, or pullet farm

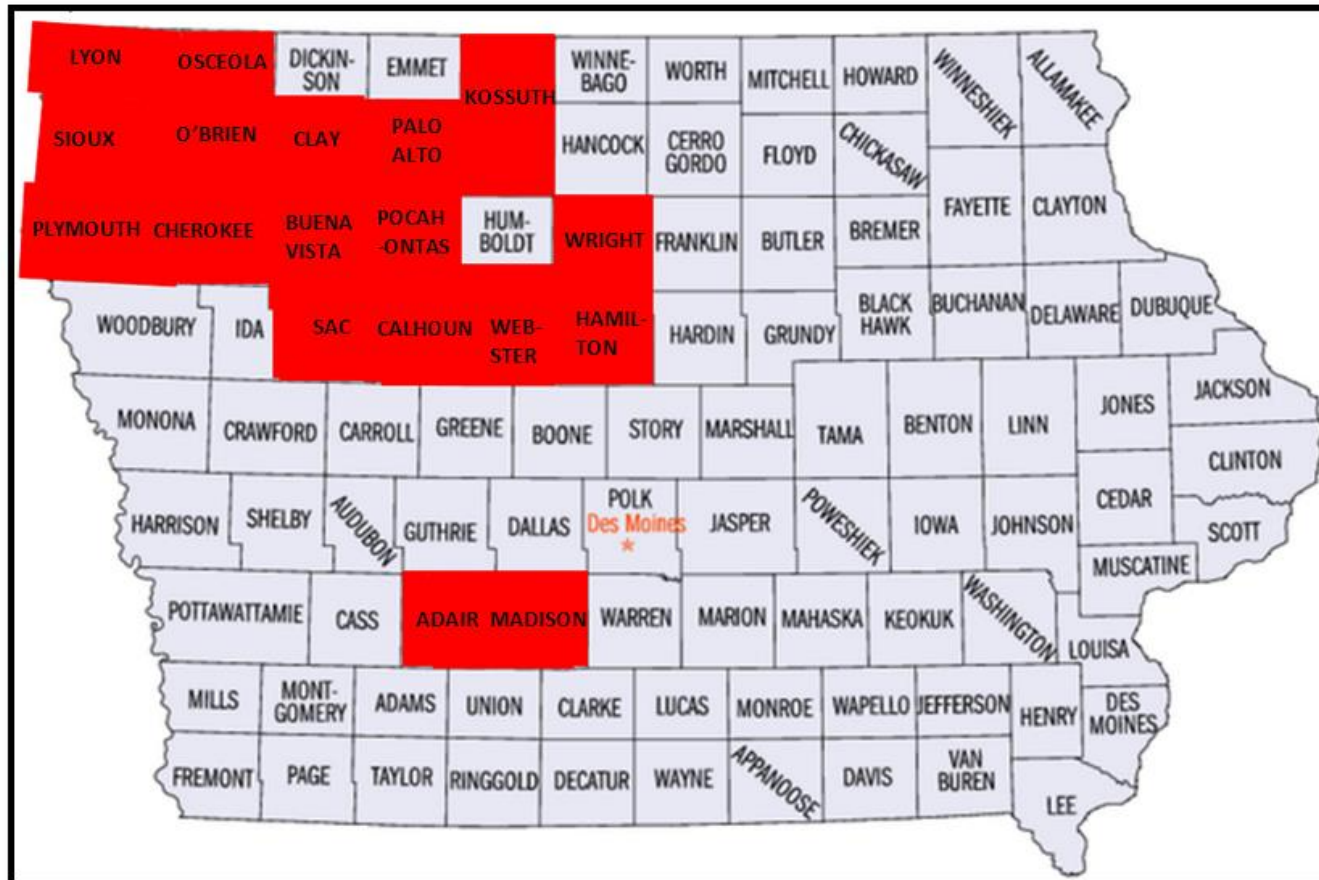


Figure 7: Distribution of highly pathogenic avian influenza (HPAI)-infected laying hen, turkey, and pullet farms in Minnesota counties during April 2015-June 2015

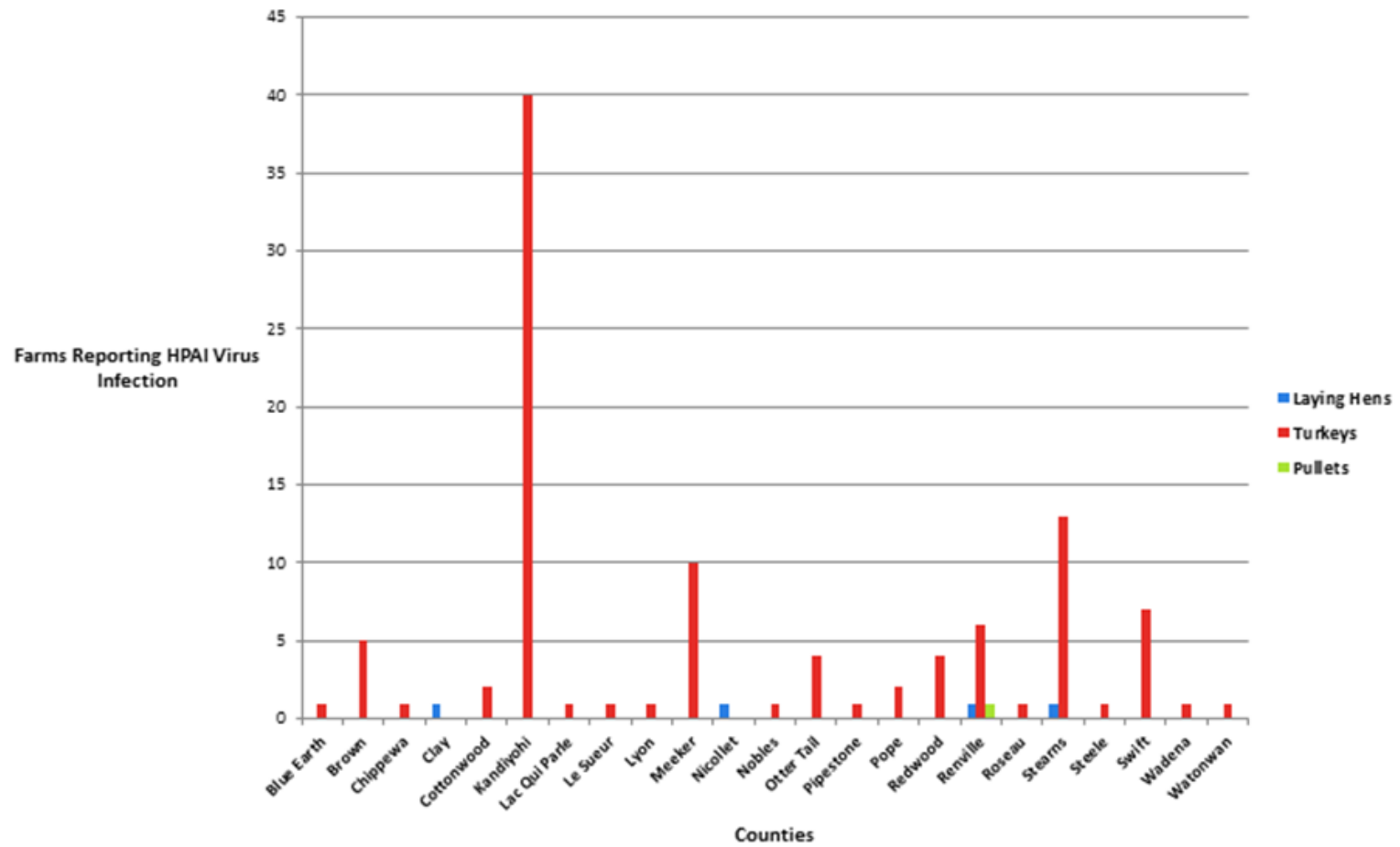


Figure 8: Distribution of highly pathogenic avian influenza (HPAI)-infected laying hen, turkey, and pullet farms in Iowa counties during April 2015-June 2015

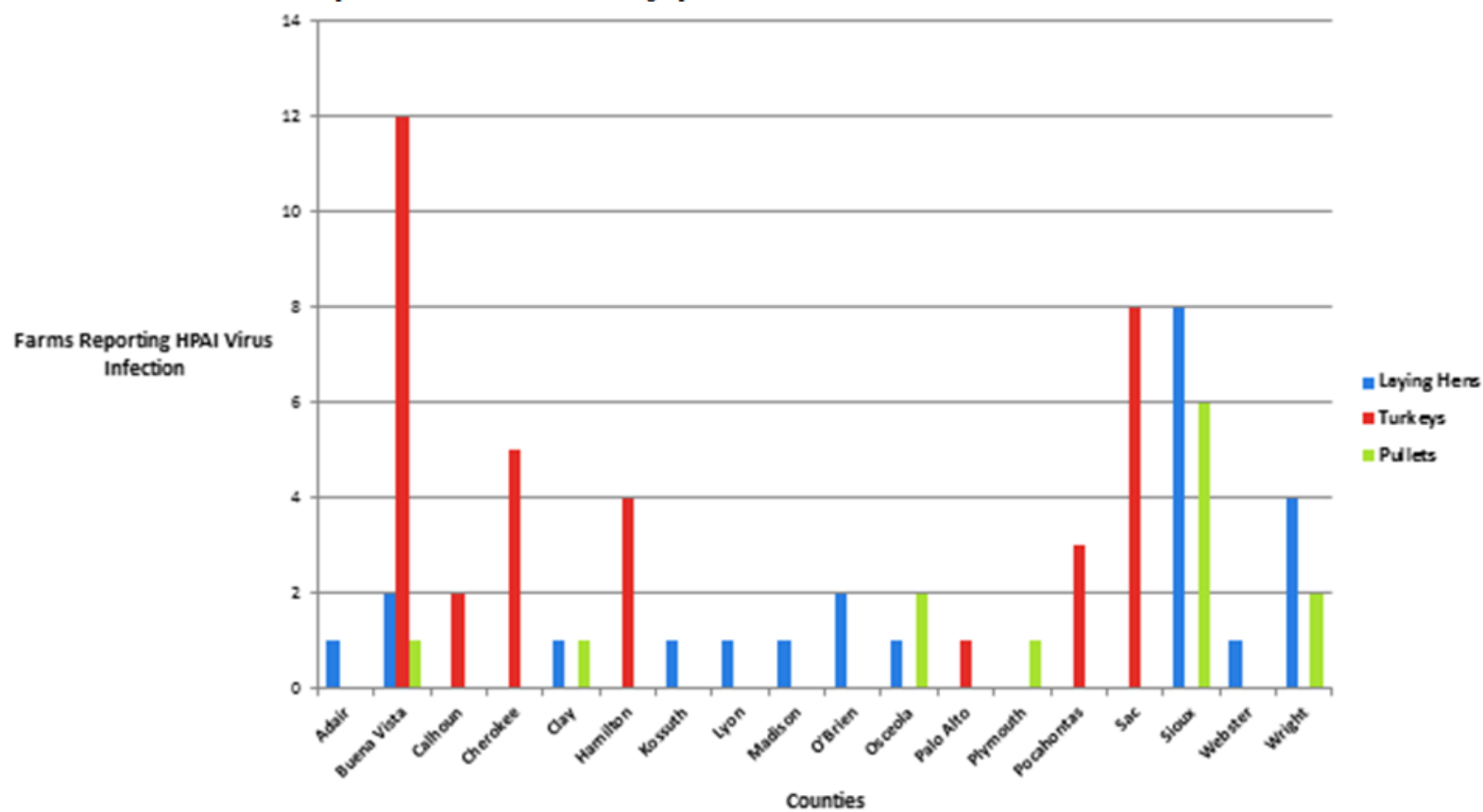
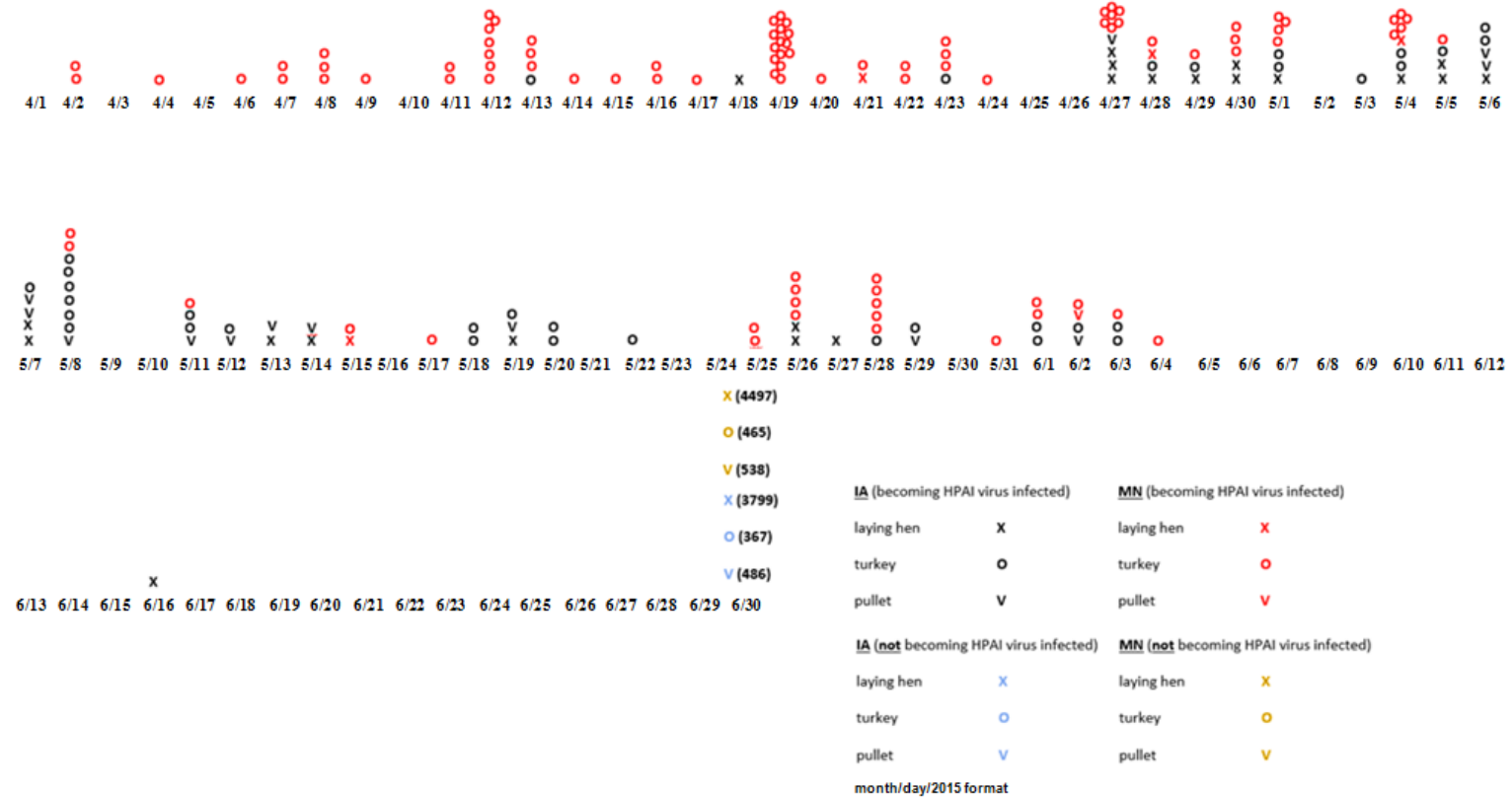


Figure 9: Days at risk of HPAI virus infection contributed by laying hen, turkey, and pullet farms during the observation period



[illegible]

A map of Iowa showing all 99 counties. The county of Sac is highlighted with a red rectangle, and the county of Buena Vista is also highlighted with a red rectangle. Other counties labeled include Lyon, Osceola, Dickinson, Emmet, Winnebago, Worth, Mitchell, Howard, Allamakee, Slick, O'Brien, Clay, Pella Alto, Kossuth, Hancock, Cedar, Grundy, Floyd, Chickasaw, Winneshiek, Keokuk, Clayton, Plymouth, Cherokee, Buena Vista, Des Moines, Hamilton, Wright, Franklin, Butler, Bremer, Fayette, Adams, Pocahontas, Woodbury, Ida, Sac, Calhoun, Webster, Hamilton, Hardin, Black Hawk, Buchanan, Delaware, Dubuque, Monona, Crawford, Carroll, Greene, Boone, Story, Marshall, Tama, Benton, Linn, Jones, Jackson, Harrison, Shelby, Guthrie, Dallas, Polk, Osage, Jasper, Pottawattamie, Cass, Adair, Madison, Warren, Marion, Mahaska, Keokuk, Muscatine, Scott, Clinton, Montgomery, Adams, Union, Clarke, Lucas, Monroe, Wapello, Jefferson, Henry, Des Moines, Fremont, Page, Taylor, Ringgold, Decatur, Wayne, Davis, Van Buren, Lee, and Harlan.

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Figure 11: Yield of corn grown for grain in Minnesota counties (USDA NASS Agriculture Counts 2015)

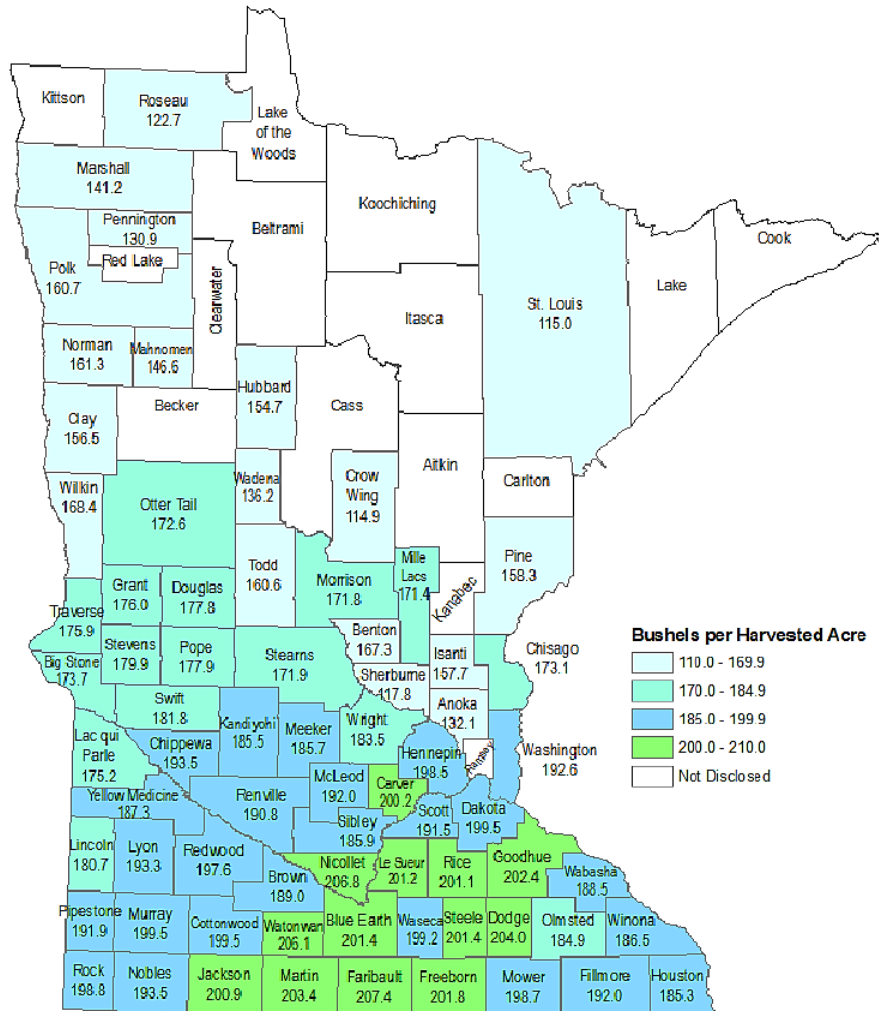


Figure 12: Yield of corn grown for grain in Iowa counties (USDA NASS Agriculture Counts 2015)

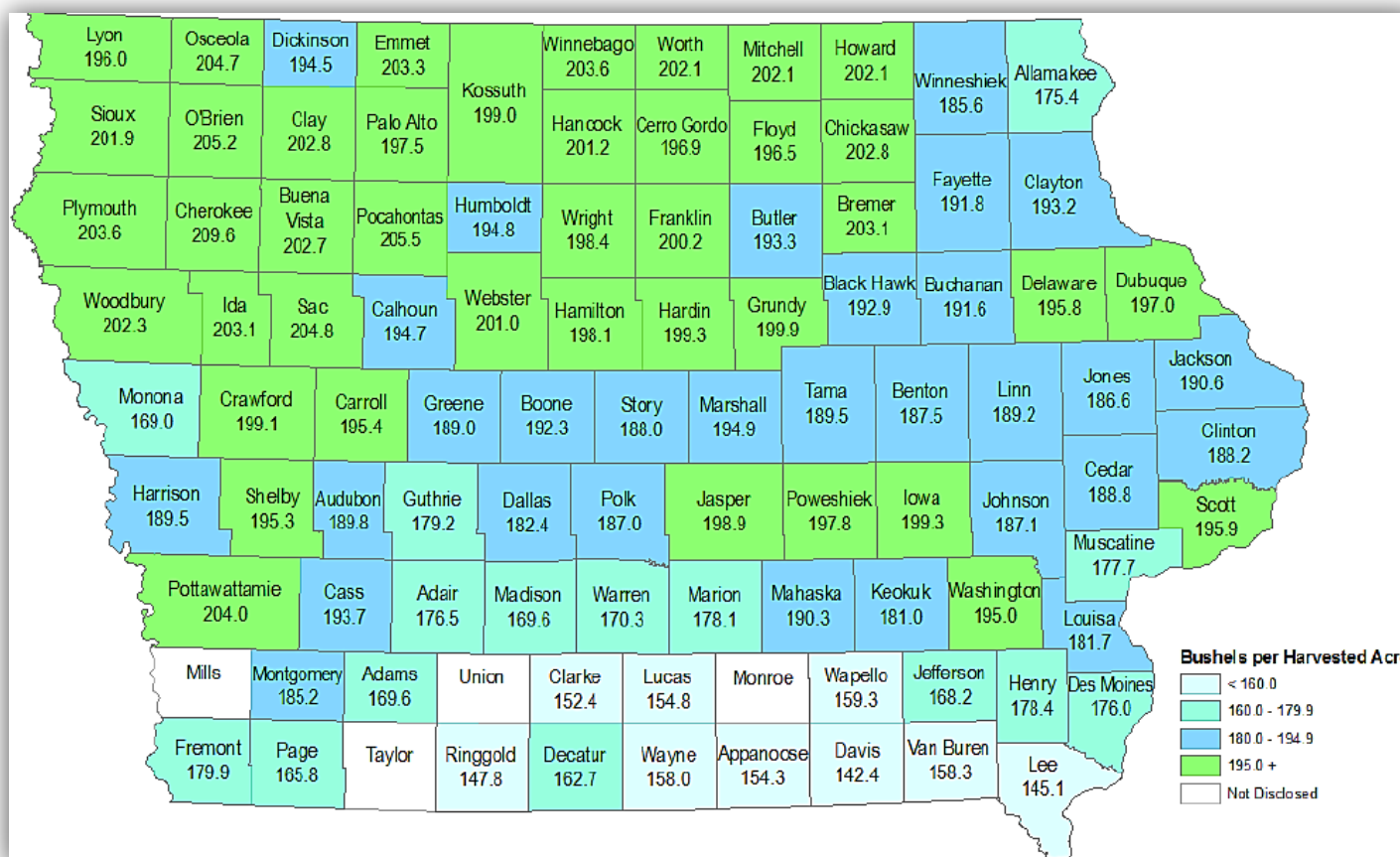


Figure 13: Farms with turkey inventory on December 31, 2012 (USDA NASS 2012 Census of Agriculture)

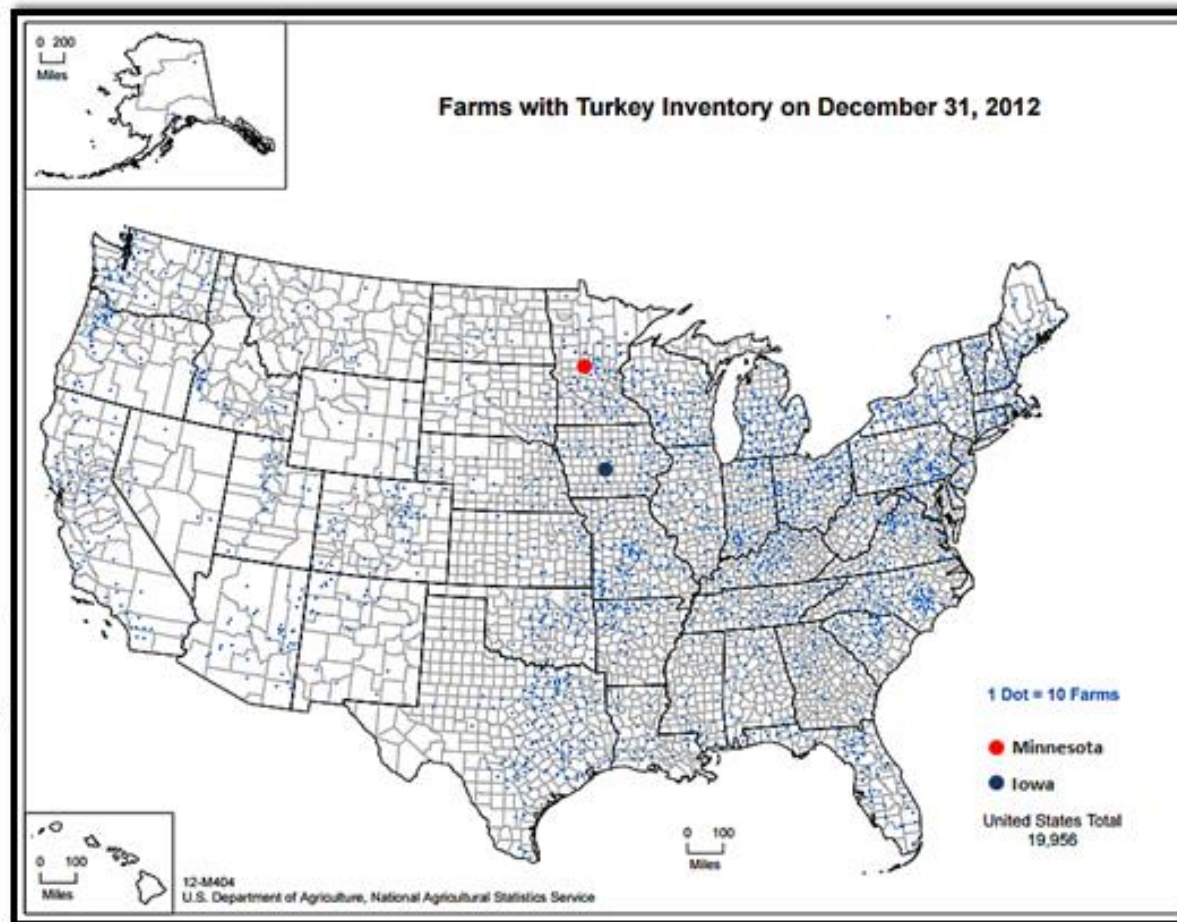


Figure 14: Farms with layer inventory on December 31, 2012 (USDA NASS 2012 Census of Agriculture)

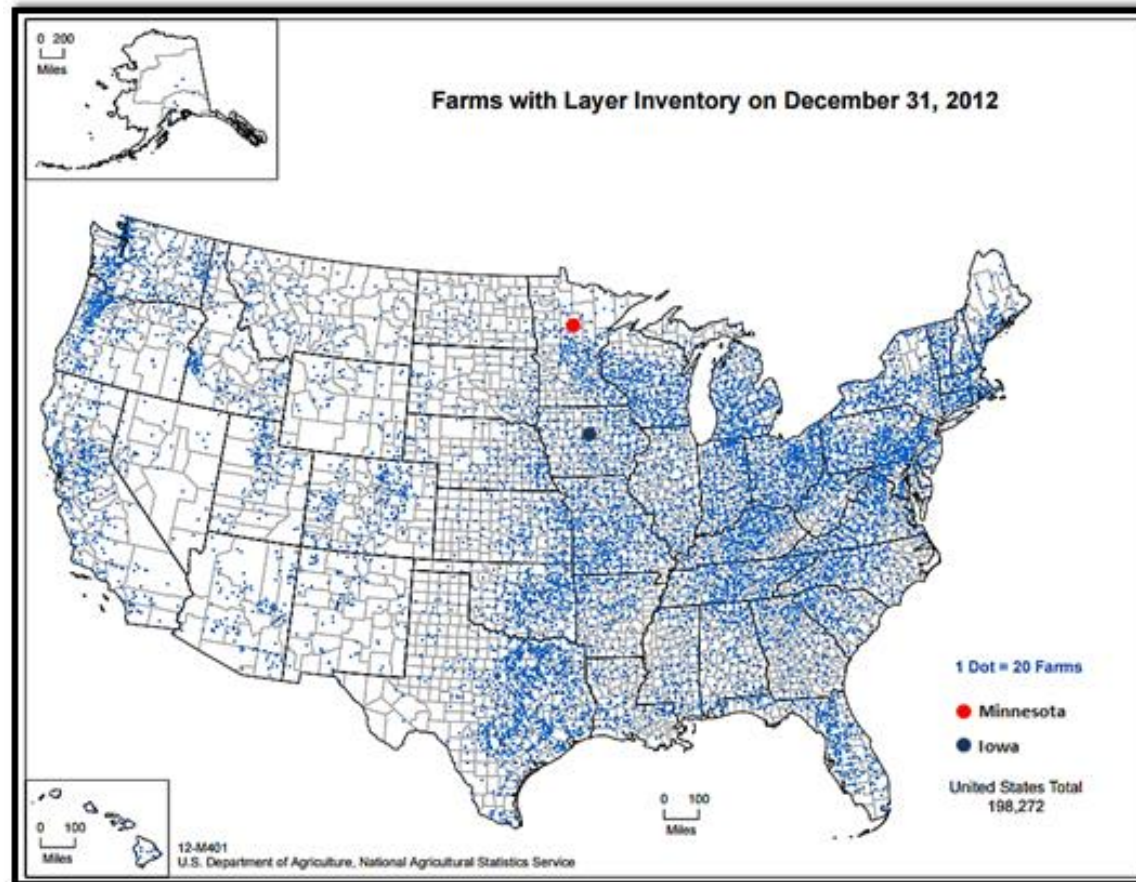
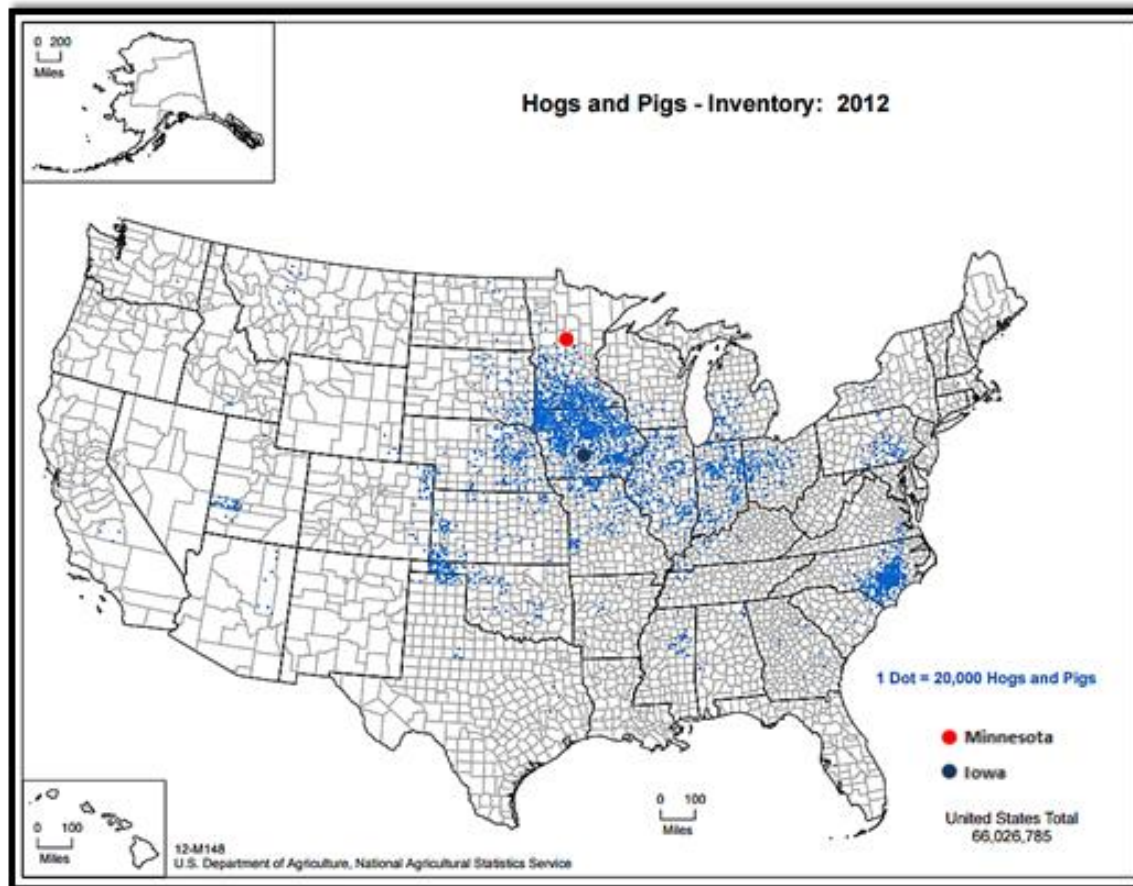


Figure 15: Hogs and pigs: Inventory 2012 (USDA NASS 2012 Census of Agriculture)



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