Exploration of risk factors contributing to the presence of influenza A virus in swine at agricultural fairs

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Influenza A virus infections occurring in exhibition swine populations at agricultural fairs during 2012 served as a source of H3N2 variant influenza A viruses transmitted to humans resulting in more than 300 documented cases. Prior to the outbreak, this investigation was initiated to identify fair-level risk factors contributing to influenza A virus infections in pigs at agricultural fairs. As part of an ongoing active surveillance program, nasal swabs and associated fair-level metadata were collected from pigs at 40 junior fair market swine shows held in Ohio during the 2012 fair season. Analyses of the data show that the adjusted odds of having influenza A virus-infected pigs at a fair were 1.27 (95% confidence interval (CI): 1.04–1.66) higher for every 20 pig increase in the size of the swine show. Additionally, four of the five fairs that hosted breeding swine shows in addition to their junior fair market swine shows had pigs test positive for influenza A virus. While the current study was limited to 40 fairs within one state, the findings provided insight for veterinary and public health officials developing mitigation strategies to decrease the intra- and inter-species transmission of influenza A virus at fairs.

Keywords: H3N2v; influenza; swine; zoonotic disease

INTRODUCTION

The comingling of swine from numerous premises with varied management practices and their interaction with large numbers of exhibitors and visitors make agricultural fairs an ideal setting for the intra- and inter-species transmission of influenza A viruses (IAVs) between swine and human populations.1,2 The frequency with which intra- and inter-species IAV transmission occurs in these settings is likely due to a myriad of factors, including but not limited to, management practices, IAV strain, and animal and/or human population immunity. Swine is a host species in which reassortment of IAV genomic segments may lead to emergent novel strains, since they are susceptible to infection from swine, human and avian influenza A viruses,3,4 For this reason, limiting the bidirectional zoonotic transmission of these viruses at agricultural fairs is important for public and animal health.

The association between human and swine influenza was reported after respiratory disease similar to the human disease was noted in pigs during the 1918 human Spanish flu pandemic.5 H1N1 IAV subsequently became established in the United States swine population with the relatedness of the swine and human viruses being established in 1931.6 For nearly 80 years, classical swine influenza H1N1 virus was the dominant endemic IAV strain in the North America swine population.7 In 1998, triple reassortant H3N2 IAVs containing polymerase basic 1 (PB1), hemagglutinin (HA) and neuraminidase (NA) gene segments from human IAV lineages, polymerase basic 2 (PB2) and polymerase acidic (PA) genes from avian lineages, and nucleoprotein (NP), matrix (M) and non-structural (NS) gene segments from swine lineages, emerged in North American swine.8 Subsequently, this lineage became established in US and Canadian swine herds and has resulted in an increased rate of genetic and antigenic change among swine-origin IAVs.9–11

Reported cases of humans contracting IAV infections directly or indirectly from pigs have been historically sporadic and these variant IAVs showed limited capability for sustained human-to-human transmission.12–14 However, the emergence of the influenza A(H1N1)pdm09 virus, a strain containing gene segments from North American and European swine lineages,15 illustrated the pandemic potential of swine lineage IAVs crossing the species barrier to humans. While A(H1N1)pdm09 rapidly spread worldwide and became endemic in the human population,16 sequencing of this virus has to date failed to elucidate any virulence or adaptation markers that would explain its human-to-human transmission efficiency, highlighting our inability to predict IAVs with pandemic potential. While the origin of the A(H1N1)pdm09 virus remains unknown, the virus was introduced into the North American swine population in 2009 and has since reassorted with other swine-origin IAVs.17,18

Epidemiological data show that zoonotic transmission of IAV from swine to humans has been documented at unprecedented levels in recent years. More than 320 human cases of infection with variant IAVs were reported to the Centers for Disease Control 2011–201219 and likely thousands more H3N2v cases went unreported during those years.20 These zoonotic IAVs contained seven genes from contemporary North American swine lineage IAV and one gene (M) derived from the H1N1pdm09 virus.21,22 The majority of the cases were epidemiologically linked to swine exposure occurring at agricultural fairs across several states.19,23–25 Within Ohio, 107 H3N2v cases documented during 2012 resulted in eleven hospitalizations and one fatality.26 We recovered IAV from exhibition swine at 10 of 40 (25%) Ohio fairs sampled during 2012. Genomic analyses of H3N2 IAV isolates recovered from pigs at one agricultural fair in the state during 2012 demonstrated
>99% nucleotide similarity to H3N2v isolates recovered from concurrent human cases, providing molecular confirmation of zoonotic IAV transmission. This record number of variant influenza A cases created the need for a ‘one health’ effort to minimize intra- and bidirectional inter-species IAV transmission at swine exhibitions. The reason to prevent IAV infections among swine at fairs is clear; however, the paucity of scientific evidence makes it difficult for veterinary officials to make sound recommendations to protect public and animal health.

In the present study, we investigate fair specific risk factors contributing to the emergence of influenza A virus in exhibition swine that could be altered to mitigate the risk of IAV transmission in these settings.

**MATERIALS AND METHODS**

As part of an ongoing active IAV surveillance project, swine nasal swabs and associated metadata about management practices were collected at 40 Ohio fairs in 2012. Molecular and microbiological assays for IAV were performed on the swabs as previously reported. Sample size was selected to provide a 95% probability of detecting IAV infection if greater than 15% of the pigs at each fair were infected. All pigs sampled in this study were from junior fair market swine shows occurring at agricultural fairs. For the outcome of interest, a fair was considered positive if viable IAV was recovered from one or more pigs at the fair.

Data collection focused on fair level variables possibly contributing to the presence or absence of IAV in the pigs at each exhibition. Junior fair shows are limited to local exhibitors approximately nine years of age through 19 years of age participating in 4-H, FFA or another youth organization, whereas open-class shows are generally open to all participants regardless of age, affiliation or residence. Classification of swine included market swine (pigs bred, raised and intended for food purposes) and breeding swine (gilts, sows and/or boars being raised for breeding purposes). Terminal swine shows are those in which all participating livestock are consigned to harvest immediately following the exhibition and partial terminal shows usually require the champion animals to be harvested following the exhibition and other pigs may or may not go to harvest.

To account for the variability of arrival and departure procedures among fairs enrolled in this study, the length of the swine exhibition was defined as the number of days between the required arrival deadline for the pigs and the day the pigs were sampled. Study team members calculated the area per pig (ft²/pig) from the recorded size of the pen and the number of pigs per pen. While on the fairgrounds, study team members also documented if there was an easily identifiable and functional hand-wash and/or hand-sanitizer stations within close proximity to the swine barn(s). These sanitation stations were used by study team members to determine if they were functional.

Additional variables included the number of pigs at the fair, number of swine exhibitors, fair attendance (number of people) and vaccine requirements, all of which were reported to the study team by the fair organizers. Fair officials also reported if there was a commingling event, such as a pre-fair animal identification or weighing session, during the weeks or months prior to the fair. Exhibition directors also reported if there were other pigs besides the exhibition swine on the fairgrounds (i.e., petting zoo, pig races, educational displays). The commercial swine inventory was retrieved from United States Department of Agriculture’s 2007 Census of Agriculture. The county human population was defined as the value reported in the 2010 US census report. Weather data were collected from the National Weather Service’s weather station nearest to each fairground.

Data were analyzed using STATA Version 11.1 (StataCorp, College Station, TX, USA). Fisher exact test was used to assess differences in proportions and the Kruskal–Wallis equality-of-populations rank test was used for continuous variables. Univariate analysis was performed to calculate unadjusted odd ratios to identify factors contributing to the presence of IAV in pigs at fairs. Exact logistic regression was used for multivariable modeling using a forward stepwise model building approach. A cutoff of \( P \leq 0.05 \) was used for inclusion in the model.

**RESULTS**

Influenza A virus was recovered from pigs at 10 of the 40 fairs included in the investigation. The presence or absence of IAV infection among the pigs at the fairs could not be associated with county population, county swine inventory and number of people attending the fair (Table 1). All IAV-positive fairs and 27 of 30 (90%) negative fairs were mixed sex (barrows and gilts) market swine exhibitions. The average space per pig at the studied fairs was 12.8 ft²/pig. Properly functioning hand-wash and/or hand-sanitizer stations were available at 25 of 40 (62.5%) fairs. Average daily mean temperature was almost 4 °C higher for fairs with IAV-positive pigs (Table 1). Pre-fair tag-in and/or weigh-in events were rather common with 23 out of 40 (57.5%) of the enrolled fairs holding one of these events. For every increase of 20 pigs at a fair, the odds ratio of IAV infection in the pigs increased by 1.27 times (Table 2).

**DISCUSSION**

The results of the current study provide the first look at fair-level risk factors associated with IAV infections in swine at agricultural fairs. While it is likely impossible to completely prevent IAV transmission at swine exhibitions, these data can be used to develop and evaluate mitigation strategies to reduce the impact of intra- and inter-species IAV infections at swine exhibitions. Just like all other agriculture biosecurity programs, mitigation strategies which are practical, user-friendly, low cost and do not dramatically alter the fair experience for exhibitors and visitors are most likely to be considered, implemented and maintained.

Not surprisingly, larger pig shows appear to be more likely to have IAV-infected pigs than smaller swine exhibitions (Table 2). Larger swine exhibitions tend to also have open-class and breeding swine shows in addition to junior market shows. While open-class shows were common among our studied fairs (16 of the 40), only 5 of the 40 (12.5%) fairs in this study had a breeding show; 4 of those 5 (80%) fairs had IAV-infected pigs at the fair. The small number of fairs with breeding shows in this study makes analysis of this risk factor problematic; however, the finding is enough to warrant concern given that breeding swine are intended to leave the exhibition and enter a herd for progeny production. This fair-to-farm movement of pigs is a disease introduction risk for the receiving herd and a potential method to disseminate IAV strains across a larger geographic area.

Previous research has shown that environmental stressors (heat, lack of space, noise) on pigs can affect the course of various diseases in commercial swine operations. The average space per pig at the studied fairs was well above 6–8 ft²/finishing pig common throughout the swine industry. The results indicate that heat stress could be a contributing factor to IAV infections in exhibition swine; however, caution must be used when interpreting this result because the vast majority of the fairs with IAV-positive pigs occurred in a 4-week period during the middle of summer. This trend of mid-summer IAV activity in Ohio’s exhibition swine was also observed in the previous three years and could be more related to animal and/or people movement between these fairs than the weather.
temperature failed to meet the selection criteria for inclusion in the final multivariable model.

While the majority of fairs had hand hygiene stations, their presence at fairs was not linked to the occurrence of IAV among pigs at the respective fairs. The large number of H3N2v infections linked to swine exposure at agricultural fairs during 2012 suggests that hand hygiene stations also had minimal impact on zoonotic transmission of IAV in these settings. However, hand hygiene stations are critically important in protecting human health by controlling zoonotic diseases transmitted via direct contact at these venues.33

One potential mitigation strategy that has been proposed is to shorten the exhibition period.34 This would limit the time for IAV to spread among susceptible pigs and decrease the time humans are exposed to IAV-infected pigs. The length of the exhibitions in the current study was similar between IAV-positive and -negative fairs.

Table 2 Adjusted odds ratios from multivariable exact logistic regression model

|                   | Adjusted odds ratio | P value | 95% CI     |
|-------------------|---------------------|---------|------------|
| Breeding show     | 21.676              | 0.005   | 2.417−∞   |
| Number of pigs    | 1.012               | 0.012   | 1.002−1.026 |

Active recruitment of fairs with more diverse management practices is needed to study the impact of a shortened swine exhibition. No matter the length of the exhibition time, the disposition of the pigs following the show must be considered. The majority of fairs in this study (65%) had terminal junior market shows. The practice of having a terminal show was not associated with decreased odds of IAV; however, sending all the pigs to harvest at the end of each fair is expected to help protect subsequent fairs by decreasing the potential for fair-to-fair spread of IAV.

Mandated vaccinations were almost non-existent with only one fair in the current study requiring the pigs be vaccinated for *Erysipelothrix rhusiopathiae* prior to the fair; no fairs required the pigs to receive IAV vaccination before arrival. Use of IAV vaccine in exhibition swine to decrease the risk of IAV infections in swine and humans has been one of the most debated topics following the H3N2v outbreak of 2012. There are currently several commercially available swine influenza vaccines in the United States, all of which are universally indicated to reduce clinical signs of disease in pigs but appear to have limited efficacy against 2012 H3N2v strains.35 Although their impact on intra-and inter-species transmission dynamics remains unclear,36 it is expected that IAV vaccines will impart at least partial immunity to circulating strains of IAV, which should decrease viral shedding.
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