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Pseudorabies in Feral Swine in the United States, 2009–2012

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ABSTRACT: Although pseudorabies virus can affect a wide range of mammalian and avian hosts, swine are the only natural hosts of the virus. The US commercial swine industry obtained pseudorabies-free status in 2004, which was important because of the economic value of domestic swine production; however, feral swine remain competent hosts and represent a constant threat for reintroducing the virus into the commercial industry. To better assess feral swine infection status, we collected 8,498 serum samples from feral swine across the United States between 1 October 2009 and 30 September 2012. Of these, 18% were antibody positive in 25 of 35 states where samples were collected, indicating that transmission risk is widespread.

Key words: Aujeszky's disease, feral swine, gB ELISA, pseudorabies, surveillance.

Pseudorabies (PRV), also referred to as Aujeszky's disease virus, is caused by Suid herpesvirus 1 (family *Herpesviridae*). Swine are the only known reservoir of the virus. Economic impacts to swine producers include high mortality in piglets, respiratory disease in juvenile and adult pigs, and abortion or stillbirths in pregnant sows (Kluge et al., 1999). Transmission primarily occurs through direct contact via the venereal route (Romero et al., 2001) or horizontal (nonsexual) transmission (Smith, 2012). Numerous other mammal species are also susceptible to infection, with the disease being highly virulent and often fatal in nonsuids.

All commercial swine (herds with adequate biosecurity measures in place to prevent contact from feral and transitional swine) in the United States were recognized as PRV free in 2004 (USDA Animal

and Plant Health Inspection Service, 2008). However, feral swine are known reservoirs of PRV and may serve as a potential source of virus reintroduction for domestic swine and other susceptible mammals. Animals that survive PRV exposure maintain a latent infection for life, leading to viral shedding and potential transmission to susceptible animals during recrudescence (Howarth, 1969).

Feral swine (*Sus scrofa*) in the United States are typically escaped domestic pigs, descendants of introduced European wild boar populations, or a hybrid of the two. They are well adapted for survival in a variety of climates and habitats, and the population is continuously expanding because of a lack of natural predators. This expansion is occurring not only into new counties in states where they are already known to exist, but also into new states. Some populations have expanded naturally, but illegal transportation and escaped feral swine from hunting preserves have contributed to increasing feral swine populations, perpetuating not only the expansion, but also the opportunities for disease transmission (Hahn et al., 2010).

The US Department of Agriculture, Wildlife Services' National Wildlife Disease Program (NWDP), coordinates a national surveillance program in feral swine with the main goal of monitoring high risk areas for introduction of classical swine fever (CSF) and other diseases with regulatory implications. Serum to test for CSF, PRV, and various other diseases is collected from swine and submitted to

laboratories across the United States proficient in testing for these diseases.

From 1 October 2009 through 30 September 2012, NWDP wildlife disease biologists collected samples from feral swine in 35 states. The majority of the samples were collected opportunistically from animals killed for wildlife damage management purposes, and a small portion of the samples was collected from hunter-killed animals. Data were recorded for each sample including animal gender, age, and GPS coordinates in decimal degrees (WGS84). Each animal was categorized as adult (≥ 1 yr), subadult (2 mo–1 yr), or juvenile (< 2 mo; Matschke 1967). Data on PRV prevalence were analyzed using logistic regression.

Once feral swine were euthanized, a blood sample was collected via cardiac puncture or orbital draw. Blood was allowed to clot and centrifuged, and sera were aliquoted into 2-mL Corning® cryovials (Corning Incorporated, Lowell, Massachusetts, USA) and labeled with a unique barcode for disease testing. Sera were stored at 4 C and shipped on ice packs to the NWDP headquarters in Fort Collins, Colorado. Samples were temporarily stored in a -80 C freezer and were batch shipped weekly. The majority of the samples were sent alternately to the Washington Animal Disease Diagnostic Laboratory in Puyallup, Washington, or the Wisconsin Veterinary Diagnostic Laboratory in Barron, Wisconsin. Samples were tested for antibodies using the PRV gB enzyme-linked immunosorbent assay (HerdCheck, IDEXX Laboratories, Inc. Westbrook, Maine, USA). Testing was considered complete regardless of the result; the primary purpose of the surveillance program was to determine the distribution of PRV in feral swine populations (presence/absence) to assess risk in domestic swine. Results from the two labs were compared using chi-square analysis and there was no difference in the proportion of antibody-positive animals reported from each laboratory ($\chi^2=2.4$, $P=0.12$).

From 1 October 2009 through 30 September 2012, 8,498 samples were collected for testing from 35 states (Fig. 1). Of these, 4,417 were female and 4,062 were male. The majority of the samples (5,458) were collected from adults, followed by subadults (2,100), and juveniles (922). Adults (prevalence=24.5%, 95% CI=22.2–24.5) were more likely to be positive than subadults (prevalence=7.9%, 95% CI=6.7–9.0) or juveniles (prevalence=5.7%, 95% CI=5.7–9.1). Males (prevalence=16.6%, 95% CI=15.5–17.8) and females (prevalence=18.9, 95% CI=17.7–20.0) were equally likely to be infected. These differences translated to a significant difference in PRV antibody prevalence between age classes ($F=145.4$, $P<0.001$), but not sex ($F=2.79$, $P=0.094$), which coincides with findings in European boar (Muller et al., 1998) and in an isolated population of feral swine (Pirtle et al., 1989). Antibodies were found in swine in 168 counties in 25 of the 35 states where samples were collected (Fig. 1).

Feral swine are persistent reservoirs of PRV in the United States (USDA Animal and Plant Health Inspection Service, 2008). Other studies have suggested that once an area is determined to be positive, it will be perpetually positive and further surveillance is unnecessary (Corn et al., 2004). Our data are relatively consistent with this pattern. For example, there were six counties (in Florida and Georgia) where PRV antibody-positive feral swine have been documented over the past 24 and 32 years respectively (Pirtle et al., 1989; van der Leek et al., 1993), and we also identified antibody-positive feral swine in these counties each year during our study; however, there were multiple counties with feral swine that were initially PRV antibody positive that yielded no positive animals in subsequent surveillance efforts. Although this might suggest a focal PRV elimination event in these counties, it more likely reflects a drop in prevalence below the detectable limits in the populations we sampled.

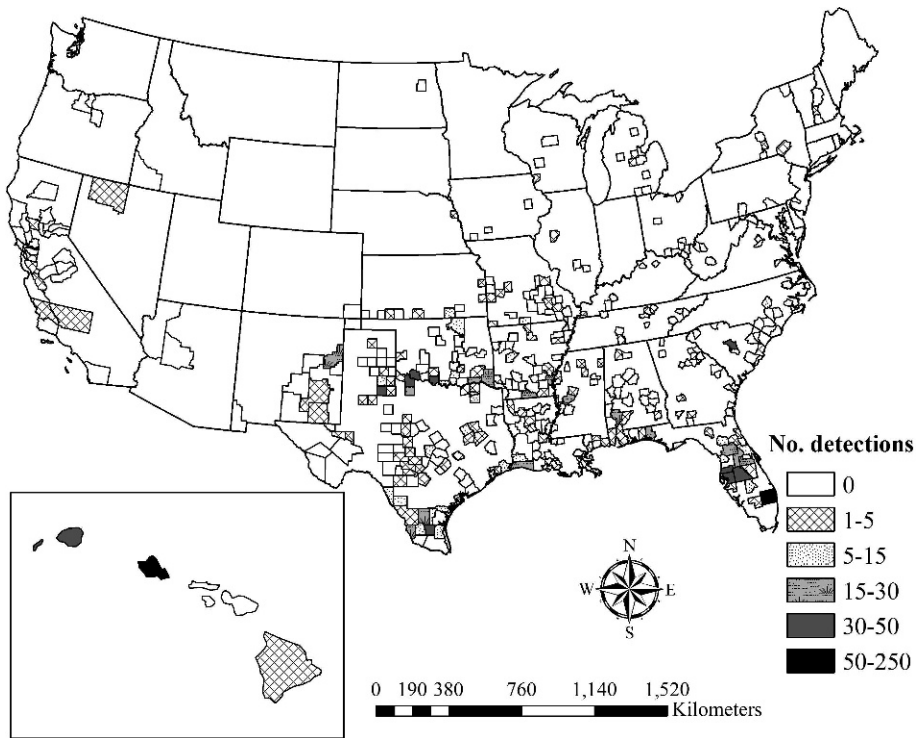


FIGURE 1. Number of pseudorabies antibody-positive detections in feral swine (*Sus scrofa*) in the United States, 2009–2012.

Because our samples were collected opportunistically, repeated sampling in the same county often occurred within a year and in subsequent years regardless of the PRV results. Detailed analysis of PRV disease dynamics in wild boars in Germany demonstrated that regional antibody prevalence can be dynamic in space and time, suggesting that repeated sampling over time is justified (Thulke et al., 2005). Repeated monitoring of feral swine in some regions might allow a similar “time-series” analysis of PRV dynamics in the United States and improve the assessment of risk to commercial swine on a local scale.

We compared counties where large sample sizes had been collected to counties with the highest antibody prevalence and there appeared to be a correlation, suggesting a threshold value of approximately 100 samples to detect a positive. However, because feral swine population

sizes and densities in counties are unknown we were unable to statistically verify this observation in the data.

The commercial swine industry is important to the United States and the gross income each year is estimated at \$16 billion (USDA National Agricultural Statistics Service, 2010). Although the primary concern in the United States is maintaining the PRV-free status of the commercial swine industry, other animals can become infected. Florida panthers (*Felis concolor coryi*) are listed as an endangered species. They are susceptible to PRV and consume feral swine as part of their diet, making exposure to infected feral swine inevitable (Glass et al., 1994). Deaths from PRV have been documented in Florida panthers (Glass et al., 1994). Although their endangered status makes susceptibility more concerning, all scavenging mammals that feed on infected

carcasses have the potential to become infected. Pseudorabies has also been documented in black bears (*Ursus americanus*; Pirtle et al., 1986; Schultze et al., 1986) and brown bears (*Ursus arctos*; Zanin et al., 1997) where infection was linked to proximity to or consumption of infected swine. As feral swine populations continue to expand and overlap with bear habitat there will likely be more reports of disease. This is especially important for species such as the Louisiana black bear (*Ursus americanus luteolus*), which is a federally listed threatened species. Hunting of feral swine is becoming popular in many states as feral swine populations increase. Disturbances related to hunting may result in animal dispersal, and with it, the probable spread of PRV infection (Muller et al., 1998). Infection of hunting dogs with feral swine PRV has been documented, and these infections are typically fatal (Cramer et al., 2011).

Our data provide an updated look at PRV exposure in feral swine from across the United States. We document widespread exposure to PRV and show evidence of infection in previously unknown locations. The expansion of feral swine populations represents a threat to the PRV-free status of commercial swine, as well as to other domestic animals and wildlife.

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