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PEER REVIEWED **ORIGINAL RESEARCH** Modelling contamination of trucks used in the shipment of pigs infected with porcine reproductive and respiratory syndrome virus

Krishna K. hakur, BVSc&AH, MS, PhD; Crawford W. Revie, BSc, MSc, PhD; Daniel Hurnik, DVM, MS; Javier Sanchez, DVM, PhD

Summary

Objectives: To quantify the likelihood that a shared truck used to ship pigs will be contaminated with porcine reproductive and respiratory syndrome (PRRS) virus at the end of a given day, and to evaluate the efficacy of cleaning and washing protocols for trucks, using a Bayesian approach.

Materials and methods: PRRS virus-infected farms, from which trucks had shipped pigs, were deemed to be the source of contamination. A quantitative stochastic model was built using farm- and animal-level PRRS prevalence data, the number of times a truck is typically shared on any given day, shipment

Resumen - Modelando la contaminación de camiones utilizados en el embarque de cerdos infectados con el virus del síndrome reproductivo y respiratorio porcino

Objetivos: Cuantificar la probabilidad de que un camión compartido, utilizado para embarcar cerdos ese contamine con el virus del síndrome reproductivo y respiratorio porcino (PRRS) al final del día, y evaluar la eficacia de los protocolos de lavado y limpieza de camiones, utilizando un acercamiento Bayesiano.

Materials y métodos: Se consideró que las granjas infectadas con el virus del PRRS, de las cuales los camiones habían embarcado cerdos, eran la fuente de la contaminación. Se construyó un modelo estocástico cuantitativo utilizando datos de la prevalencia del PRRS a nivel animal y de granja, el número de veces que un camión se comparte típicamente en un size, travel time between farms, and the efficacy of three diferent cleaning and disinfection procedures.

Results: The model predicted a median probability of 0.525 that a truck would be contaminated at the end of any given day, without considering the number of previous shipments made by that truck or whether or not it had been washed and disinfected between shipments. Truck washing alone resulted in a negligible decrease in probability that a truck would be contaminated, while washing and disinfection followed by drying had the highest impact, with a greater than 99% reduction in probability of contamination.

día, el tamaño del embarque, tiempo del viaje entre granjas, y la eficacia de tres procedimientos diferentes de limpieza y desinfección.

Resultados: El modelo predijo una probabilidad mediana de 0.525 para que un camión se contamine al final de un día dado, sin considerar el número de embarques previos hechos por el camión o si se había o no lavado y desinfectado entre embarques. El sólo lavado del camión resultó en una disminución insignificante en la probabilidad de que un camión se contaminara, mientras que el lavado y la desinfección seguidos de secado, tuvieron el impacto más alto, con una reducción mayor al 99% en la probabilidad de contaminación.

Implicaciones: Los hallazgos de este estudio sugieren que bajo las prácticas actuales de bioseguridad, existe un riesgo substancial para la propagación del virus del PRRS debido al

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Implications: Findings of this study suggest that under current biosecurity practices, a substantial risk exists for the spread of PRRS virus due to truck sharing. This model could also be utilized in understanding the risk of truck sharing on the spread of other swine diseases (such as porcine epidemic diarrhea) where transportation is believed to spread the virus.

Keywords: swine, Bayesian, porcine reproductive and respiratory syndrome virus, truck sharing, shipment

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hecho de compartir el camión. Este modelo, también podría ser utilizado, para entender el riesgo de compartir camiones en la propagación de otras enfermedades porcinas (tales como la diarrea epidémica porcina) donde se cree que el transporte propaga el virus.

Résumé - Modélisation de la contamination des camions utilisés dans le transport de porcs infectés par le virus du syndrome reproducteur et respiratoire porcin

Objectifs: Quantifier la possibilité qu'un camion partagé utilisé pour le transport de porcs sera contaminé par le virus du syndrome reproducteur et respiratoire porcin (SRRP) à la fin d'une journée donnée, et évaluer l'efficacité des protocoles de nettoyage et de désinfection des camions, par une approche bayésienne.

Matériels et méthodes: Les fermes infectées par le virus du SRRP à partir desquelles des camions furent utilisés pour expédier des porcs étaient considérées comme étant la source de la contamination. Un modèle stochastique quantitatif a été construit en utilisant les données de prévalence du SRRP au niveau de la ferme et au niveau des animaux, le nombre de fois typique qu'un camion était partagé à chaque jour, la taille de l'expédition, le temps de transit entre les

fermes, et l'efficacité de trois procédures différentes de nettoyage et désinfection.

Résultats: Le modèle a prédit une probabilité médiane de 0,525 qu'un camion serait contaminé à la fin d'une journée, sans prendre en considération le nombre de transports précédents efectués par ce camion ou s'il avait été ou non lavé et désinfecté entre les transports. Uniquement un lavage du camion a entrainé une diminution négligeable de la probabilité qu'un camion serait contaminé, alors que le lavage et la désinfection suivis par un séchage avaient le plus grand impact avec une réduction de plus de 99% de la probabilité de contamination.

Implications: Les résultats de cette étude suggèrent qu'en fonction des pratiques de biosécurité actuelles, un risque substantiel existe pour la dissémination du virus du SRRP dû au partage de camions. Ce modèle pourrait également être utilisé pour comprendre le risque de partage de camions dans la dissémination d'autres maladies porcines (telle que la diarrhée épidémique porcine) pour lesquelles le transport est considéré comme pouvant disséminer le virus.

 $\sum \begin{array}{c} \text{orcine reproductive and respiratory} \\ \text{tory syndrome (PRRS) is a magic} \\ \text{viral disease of swine with devas} \\ \text{ing economic consequences to the swi} \end{array}$ tory syndrome (PRRS) is a major viral disease of swine with devastating economic consequences to the swine industry.1,2 Pigs of all ages are susceptible to PRRS virus, with highly variable clinical manifestations in diferent age groups of infected pigs, ranging from abortion, anorexia, and pyrexia in pregnant sows to respiratory signs, high mortality, and poor growth in young pigs.³ Several direct and indirect mechanisms have been identified for the spread of PRRS virus from one farm to another. Direct contact routes include movement of infected animals and use of contaminated semen. Indirect mechanisms involve sharing shipment trucks between farms^{4,5} and sharing of equipment and other fomites.6-8 Aerosol transmission has also been reported, 9,10 and some studies have implicated the role of insects, such as mosquitoes and house flies, in mechanical transmission of the virus.^{11,12}

he swine industry in North America has become increasingly specialized and integrated, with the adoption of three-site production systems that require regular movement of pigs between sites.13 Canadian swine producers and experts in the swine industry have been concerned about the role of shared trucks in the farm-to-farm spread

of PRRS virus.¹⁴ In recent years, network analysis has elucidated contact patterns among animal holdings in specific livestock industries.15-18 A number of recent swine movement analysis studies in Denmark and France have identified the importance of shipment vehicles as a means to spread infectious agents between farms that are otherwise not directly connected.17,19,20 In a recent study characterizing swine movement in four Canadian regions, ¹⁸ patterns of truck-sharing between farms, similar to those described in the United Kingdom,¹⁷ France, 20 and Denmark, 19 were identified, with one truck, on average, being shared among four diferent farms. Similarly, for more than 50% of all shipments on any particular day, the same truck had been used in at least one additional shipment from a different farm.¹⁸ Thus, any inadequate cleaning and disinfection of vehicles is likely to increase the risk of spread of infectious agents, as has been implicated in the recent spread of porcine epidemic diarrhea (PED) virus across North America.²¹

he role of shipment vehicles in the spread of PRRS virus to susceptible pigs has also been demonstrated through experimental studies in which sentinel pigs became infected after being housed in an artificially contaminated trailer and in trailers that housed experimentally infected pigs.⁴ Similarly, mechanical transmission of PRRS virus via transportation during cold and warm weather has been documented by the same authors.22,23 Trucks contaminated with PRRS virus require rigorous cleaning, disinfection, and drying to eliminate the virus. 4.24 he Canadian Swine Health Board has developed protocols to wash, disinfect, and dry such transport vehicles.²⁵ However, anecdotal evidence indicates a lack of consistency in the application of these standardized protocols, with some trucks being cleaned by washing only, while others are washed and disinfected, and others undergo the full protocol of washing, disinfection, and overnight drying. In these experiments, Dee et al 4 also evaluated cleaning and disinfection protocols. PRRS virus from the trailers was detected in all combinations of cleaning and disinfection treatments except when bedding removal, washing, disinfecting, and drying were combined. Washing and fumigation with glutaraldehyde-quaternary ammonium chloride or washing and disinfection plus overnight drying were efective treatments.²⁴

A Bayesian approach was selected for this study, as it supports a combination of diferent sources of information and the propagation of uncertainty in the model.26 It also allows the assumption of conditional dependence between nodes required by classical risk assessment to be relaxed, supporting the estimation of joint probability distributions at nodes that are conditionally independent, through the use of Bayesian networks.27 A Bayesian network is a probabilistic graphical model representing a network of nodes connected by directed links, where nodes represent a set of random variables, and the links, dependencies between these nodes. Bayesian networks have been used in veterinary epidemiology to aid in disease diagnosis²⁸⁻³⁰ or to study associations between biosecurity practices and disease outbreak.31,32

he objectives of the analyses described here were to use a Bayesian approach to quantify the likelihood that a truck used for a shipment of pigs would be contaminated with PRRS virus at the end of any given day (as well as on subsequent days), and to evaluate the efficacy of cleaning and disinfection protocols in eliminating the virus from these trucks. The model estimates the probability that a truck will be contaminated with PRRS virus after it has been used by a number of farms on any given day, and provides estimates on the likelihood that the truck will remain contaminated on subsequent days. In addition, it provides insights into the likelihood that the PRRS virus will be eliminated from the trucks ater one of a number of cleaning and disinfection protocols has been applied.

Materials and methods

Truck-use pattern for transportation of pigs between swine farms, informed by pig traceability data from a pilot study in Canada, 18 suggests that a given truck may be used by two or more farms on any given day, and may or may not be cleaned between shipments. A schematic representation of the Bayesian model is presented in Figure 1. he baseline model estimates the probability that a truck "i" will be contaminated with PRRS virus at the end of Day 1, given it visited "j" farms on that day, and that at least one of those farms was infected with PRRS virus. It also estimates whether the truck had sufficient viral load to make it infectious, which is determined by the travel time of the truck during the shipment, the number of pigs in the shipment (ie, shipment size), the animal-level prevalence of the virus, and the probability of animals

Figure 1: Schematic representation of the Bayesian network used to estimate the likelihood that shipment trucks would be contaminated with porcine reproductive and respiratory syndrome (PRRS) virus and to evaluate the efficacy of various cleaning and disinfecting protocols for removal of PRRS virus from contaminated trucks. Fixed nodes are shown in rectangles, with stochastic nodes in ellipsoids. he diamond-shaped node represents an outcome that is estimated by the model. Nodes in yellow represent prior information. The model represents a truck_i, that can have visits_j (two, three, four, or more) on a day and can ship animals from production types_k (farrowing, nursery, or finishing) of swine farms and can be cleaned by using protocols_l (wash; wash and disinfect; or wash, disinfect, and dry).

shedding in the shipment group. The probability of shedding animals depends on the production type "k" of the infected farm.

he baseline model incorporated one of the three "l" cleaning and disinfection protocols and evaluated the efficacy of each of these protocols in eliminating the PRRS virus from contaminated trucks. This risk assessment considered the following five nodes as influencing the probability that a truck would be contaminated with PRRS virus at the end of a given day: farm-level prevalence of PRRS virus (proportion of PRRS farms with at least one PRRS-positive animal), number of farms using the same truck on that day, number of animals shipped on the truck, animal-level prevalence of PRRS virus in the group of shipped pigs, and proportion of animals shedding the virus in the group, which in turn depends on the stage of growth of the

pigs and the time of travel between two farms. A detailed description of the nodes is presented in Table 1, with a summary of the underlying assumptions, process models, and associated input values. Finally, we estimated the probability that a truck "i" would be contaminated with PRRS virus by multiplying the probability that at least one of the farms "j" that it had visited was infected with PRRS virus, the probability that the truck had more animals in that shipment than Min_{anik} (minimum number of animals required on a truck to have enough infectious animals to contaminate the truck), and the probability that the travel time was more than 2 hours.

Data description

Data used in this study were obtained from the literature^{4,24} and from a pilot pig traceability study carried out in four Canadian regions and described elsewhere.18 As the Bayesian approach is useful in combining information from several sources, and since most of the data used in the study are based on published literature or experts' judgement, we considered these data as prior information for the model.

For this study, we assumed that a truck was free of PRRS virus when it was used for the first time on Day 1. In assessment of the perpetuation of risk on Day 2 and subsequent days, we did not consider any new sources of infection for that truck, such that all farms visited by the truck after the first day were assumed to be clean. On the basis of the experts' judgment, we assumed the farmlevel prevalence (F prev) of PRRS virus to be 50%. One co-author (DH, professor of swine health management) and an external

Table 1: List of nodes and parameters, process models, prior distributions, and observed data with source and references used to estimate the probability that a truck will be contaminated with porcine reproductive and respiratory syndrome virus (PRRSV) at the end of a working day

Table 1: Continued

† For shipment size a triangular distribution was simulated using two uniform distributions [Uniform (min/2, max/2) + Uniform (min/2, max/2)] in OpenBUGS with minimum and maximum as 5th and 95th percentiles of shipment size.

For travel time, a triangular distribution was simulated using two uniform distributions [Uniform (min/2, max/2) + Uniform (min/2, max/2)].

§ N prot = No. of clean trucks after wash, wash and disinfection, or wash, disinfection, and dry.

 \blacksquare N w = Total no. of trucks washed.

†† N wd = Total no. of trucks washed and disinfected.

‡‡ N wdd = Total no. of trucks washed, disinfected, and dried.

§§ Jefreys priors.

NA = not applicable.

expert (Dr Zvonimir Poljak, an associate professor of veterinary epidemiology with a research focus on swine diseases), who have extensive expertise in swine production and management across Canada, provided input to estimate some of the parameter values. Using the farm-level prevalence of the virus, we estimated the number of farms that might be infected with PRRS virus (of the farms visited by the truck on any day), as well as the probability that at least one of the visited farms was infected with PRRS virus. A detailed description of the estimation procedure for the values and parameters of each node is provided in the supplementary material.

Models

Two sets of models were evaluated to estimate the likelihood that the trucks shared between farms for the shipment of pigs were contaminated with PRRS virus. First, a baseline model was simulated that did not involve any cleaning or disinfection protocol being applied to the truck, which resulted in an estimation of the "baseline" probability that the truck would be contaminated at the

end of Day 1. The baseline model was then extended to incorporate decay of the virus over time under two diferent seasonal settings: warmer months (assuming an ambient temperature of approximately 22°C) and colder months (which assumed an ambient temperature of approximately 4°C or less) to assess the probability that the truck would remain contaminated on subsequent days under these conditions.

Evaluation of cleaning and disinfection protocols

he baseline model was extended to include three cleaning and disinfection protocols that have been assessed previously in terms of their efectiveness in eliminating the PRRS virus from contaminated trucks. Data from Dee et al studies^{4,24} (summarised in Table 1) were used for each of the three cleaning protocols to assess their efectiveness in reducing the probability that a truck used for shipments of pigs would remain contaminated with PRRS virus. Since these data are from a small number of replication experiments, non-informative Jefreys priors were used.

As these priors depend upon the process model33 for these nodes, beta (0.5, 0.5) was used in order to avoid a large influence of these data on the posterior estimates.

Scenarios

A total of 21 scenarios were constructed and analysed (Table 2). A subset of 12 of those scenarios did not include any cleaning or disinfection control measures, while the other nine scenarios evaluated the efficacy of each of the three cleaning and disinfection protocols. For scenarios without cleaning and disinfection protocols, the risks for trucks used by two, three, and four or more farms were evaluated, and the combined risk for a "random" truck, for which the number of farms previously visited on that day was unknown, was estimated. In addition, the production type of the initially infected farm visited by the truck was included in these scenarios. Similarly, for scenarios with cleaning and disinfection protocols, the probability that a random truck would still remain contaminated ater visiting any of the three production farm types, and would

have been cleaned by one of the three cleaning protocols, was estimated.

Stochastic model

To quantify the probability that a truck used by a number of farms on a given day would be contaminated with PRRS virus at the end of the day, a stochastic model was developed (code attached in supplementary material) in OpenBUGS 3.2.2.34 A total of 150,000 iterations, with a burn-in period of 30,000, were obtained ater initializing the model with three chains. The convergence, diagnostic analyses, and summary of all posterior distributions were computed in R using the CODA package.³⁵ The convergence of the Markov Chain Monte Carlo (MCMC) model was assessed both visually, using the history and autocorrelation plots, and formally, using the Brooks-Gelman-Rubin diagnostic,36 which provided an estimate of the shrinkage or scale reduction factor for each of the nodes and scenarios. The distribution of the scale reduction factors (median

and 97.5% upper bounds) was plotted to visually assess convergence. Once the model converged, the efective sample size was estimated by running the model for a sufficient number of iterations such that the MCMC error became less than 5% of the posterior standard deviation for monitored nodes. The median and 95% credible interval (CrI) are reported, along with the mean and standard deviations for the scenarios described above, and for each stochastic node.

Sensitivity analysis

A sensitivity analysis was performed for one scenario (f_i) , where the truck was used by an unknown number of farms and carried finishing pigs, without the application of any cleaning and disinfection measures. The aim was to evaluate and identify possible scenarios that could lead to a significant decrease or increase in the final probability. he percentage change in the mean probability of contamination was compared to the original scenario. These changes were

assessed using farm-level prevalence (F prev) values for PRRS of 10%, 30%, and 70%; animal-level prevalence (A prev) values of 10%, 30%, 50%, and 100%; and animalshedding prevalence (Shed ani) of 10%, 30%, 50%, and 90%. The sensitivity of the model was also evaluated by changing the minimum number of infectious animals required to contaminate a truck (N ani) from two to four and eight.

Results

The median probabilities (and 95th percentile of the distribution) of a truck remaining contaminated with PRRS virus at the end of Day 1 for scenarios with and without the application of various cleaning and disinfection measures are presented in Figure 2. The median and 95% CrI, along with mean and standard deviation for all the parameters used in the model and for all the scenarios, are summarised in the supplementary material, Table S1. The median probability that a truck would be contaminated with PRRS

Table 2: Scenarios created to evaluate the probability that a truck will be contaminated with porcine reproductive and respiratory syndrome virus at the end of a working day

W = truck washed; wd = truck washed and disinfected; wdd = truck washed, disinfected, and dried; fa = truck used by farrowing farms; $nu =$ truck used by nursery farms; and $fi =$ truck used by finishing farms.

virus at the end of Day 1, when it was used by three farms or more, was 1 irrespective of the production type of the farm that used that truck. However, the median probability for trucks used by only two farms was 0. In the case of a "random" truck (ie, one for which the number of times it had been used for transportation during the first day was not specified), the median probability was 0.529 when the source of infection was a finishing farm. The median probability of a truck remaining contaminated did not difer much for truck use across the three diferent production types for these scenarios.

When decay of the virus over time was incorporated in the model, the outputs suggested that the median probability that a truck would still remain contaminated ater 15 or 30 hours of use during warmer months, from a contamination acquired on Day 1 and without visiting any other infectious farms on subsequent days, was not much diferent from the probability on Day 1 (Table 3). The median probability decreased by 0.049 and 0.051 (approximately 10%) in the 15 hours of truck use subsequent to Day 1, when the truck had been used by either a nursery or a finishing farm, respectively, and by 0.063 (12.5%) when it had been used by a farrowing farm, compared to Day 1. For the next 30 hours after Day 1, the median probability of contamination decreased by 0.218 for nursery farms and by 0.211 for finishing farms, compared to Day 1. Similarly, median probability for truck contamination ater 30 hours of truck use reduced to 0 when the source of infection was a farrowing farm. For colder months, the virus can remain viable for approximately 112 hours,³⁷ so once contaminated on Day 1, the trucks were expected to remain contaminated for approximately 5 additional days.

With respect to the three cleaning protocols evaluated in this study, washing alone reduced the median probability of a truck remaining contaminated by 0.011 (for example, the probability for $fi = 0.525$ decreased to 0.514 with washing), while washing with disinfection decreased the median probability by 0.346 (approximately 66%). However, washing and disinfection followed by overnight drying had by far the highest impact, lowering the median probability of contamination by more than 99%, to approximately 0.002, irrespective of the production type for which the truck had been used (Table S2 of supplementary material). The distributions of probabilities associated with a truck remaining contaminated ater the application of each of the three cleaning and disinfection protocols, for the scenario involving finishing farms, are presented in

Figure 2: Median probabilities (with 95th percentiles, p95) for contamination of trucks with porcine reproductive and respiratory syndrome virus (PRRS virus) at the end of Day 1, for several scenarios, depending on the number of times the trucks were shared (three, four, or more) and the production type of the PRRS-virus-infected farm, (left) without cleaning and disinfection of trucks and (right) with application of one of the three cleaning protocols evaluated in the study. Only representative scenarios are presented (supplementary material is available [Table S1] for median probabilities for other scenarios). Fi = finishing farm; w = washing; wd = washing and disinfecting; wdd = washing, disinfecting, and drying.

Figure 3. Similar distributions for scenarios without cleaning and disinfection protocols could not be obtained due to the parametrizing of nodes used in those scenarios with step function, which did not allow for the propagation of uncertainty across these nodes or for the scenarios evaluated. However, we would expect those scenarios to have similar distributions to that of the "fi w" scenario.

Finally, outputs from the sensitivity analyses suggested that the highest percentage changes (100% decrease for each scenario evaluated) were observed for large decreases in farm-level or animal-level prevalence of PRRS virus and for a large decrease in the probability of shedding animals in the shipment (when either one of these was decreased to 10%). However, only a small increase or decrease in the median probabilities was observed for a smaller increase or decrease in each of the parameters evaluated (farm-level prevalence, animal-level prevalence, and the probability of shedding animals in any particular shipment (Figure 4 and supplementary material, Table S2.) Similarly, large increases

 $(2\times$ and $4\times)$ in the minimum number of infectious animals (N_{ani}) required to contaminate the truck with PRRS virus resulted in only a small decrease (approximately 10% and 40%, respectively) in the median probability of contamination.

he MCMC error was less than 5% of the posterior standard deviation for all of the reported scenarios and nodes, which suggested that the model had been run for a sufficient number of iterations, and 40,000 iterations with a burn-in period of 10,000 for each chain was sufficient to allow the models to converge with a sufficient sample size for posterior inference. The scale-reduction factor was less than 1.05 for all nodes and scenarios evaluated, indicating that the model converged. The shrinkage plots, showing the evolution of the scale reduction factor with an increase in the number of iterations, also suggest that the MCMC models converged after approximately an initial 4000 iterations, following the burn-in period of 10,000 iterations, for most nodes.

Table 3: Probability that a truck will remain contaminated with porcine reproductive and respiratory syndrome virus in subsequent time periods during warmer months

 $Fa = true$ k used by farrowing farms; nu = truck used by nursery farms; and $fi = true$ k used by finishing farms.

Discussion

his analysis evaluated the risk for contamination with PRRS virus of trucks involved in the transportation of pigs. To do so, a baseline model was first developed to assess the likelihood that trucks used for shipment of pigs will become contaminated with PRRS virus and remain so at the end of Day 1. The model was extended to explore a number of possible scenarios, including variations in the number of times a truck was used in a day, the farm- and animal-level prevalence of PRRS virus, the size of the shipment on a truck, the probability of shedding animals in the shipment, and the period of travel involved. The model was extended to quantify the probability that the truck would remain contaminated on subsequent days once it became contaminated, without visiting any other infected farms, by including decay of the virus over time in the model. We also attempted to evaluate the efficacy of commonly-used cleaning and disinfection protocols in eliminating this virus from contaminated trucks.

On the basis of this model, the estimated probability of a truck being contaminated at the end of a day increased substantially with an increase in the number of visits the truck made on a given day. However, there were no major diferences in the probabilities for scenarios when the truck was used by farrowing, nursery, or finishing farms. he two parameters that were diferent in the model among the three production types were shedding percentage and shipment size. The sensitivity analysis suggested that the model was less inluenced by changes in shedding percentage, unless it was a very large change, and that above a certain threshold for this parameter, the model behaved similarly. This explains why very limited diferences in risk were observed among the three production types.

While separate shipment size distributions were specified for the three production types, in most cases the shipment size was very large, typically large enough to have the minimum number of animals required to characterize the trucks as being contaminated. Thus, this parameter also had little impact in terms of overall diferential risk among the three production types.

he model suggested that the virus would be eliminated from only a very small proportion of trucks by simply washing the vehicle, while washing followed by disinfection should clean the virus from just over half of the trucks. Washing and disinfection, followed by overnight drying, had the highest impact, resulting in the removal of PRRS virus from a large majority of contaminated trucks. One possible explanation for the high efficacy of this protocol may be as follows. Washing alone can reduce the amount of debris and organic matter but cannot eliminate the virus, while washing followed by disinfection can be useful when the surfaces are free of organic matter. However, the addition of drying can eliminate the virus from contaminated surfaces by eliminating the residual virus that persists ater washing and disinfection has occurred.38 Findings from our study are in slight contrast with those observed in the experimental studies.5,24 In the experimental study, washing had no efect at all, and washing and disinfection were efective in approximately a quarter of replications, while washing, disinfection, and drying resulted in the elimination of the virus in all replications. The differences observed in the current study were likely due to the introduction of uncertainty and stochasticity into the model.

Finally, the model suggested that, during warmer months, a slight decrease may occur in the probability that the trucks will be contaminated on the following day, as some trucks may become decontaminated the following day simply due to viral decay. How-

ever, most trucks that are contaminated on Day 1 will remain contaminated for at least 30 hours. Again, this finding was associated with shipment size, as most of the trucks, due to large shipment sizes, qualified to have infectious and shedding animals sufficient to maintain the contamination for the next few days. In colder months, when the virus can survive much longer,³⁷ a truck will tend to remain contaminated for approximately 5 days once contaminated. Cleaning and disinfection of trucks to eliminate PRRS virus is crucial during winter months, 22,39 when the virus exhibits increased survival. However, our study suggests that cleaning and disinfection should not be ignored during the warmer months, as the likelihood that trucks will remain contaminated for a number of days following shipment from an infected farm is substantial.

In the present study, the viral load on trucks could not be quantified because data on the amount of PRRS virus that is typically shed were not available. Instead, trucks were classified in terms of whether they were likely to have sufficient viral load to be able to transmit the infection, on the basis of work by Dee et al, 4 using shipment size as a proxy for viral load. Shipment size was linked to PRRS viral load on the trucks in terms of a dose-response relationship, which further afected the time that the truck would likely remain infected with the virus. Even with decay of the virus over time, trucks that carry larger shipments from infected farms can remain contaminated for several subsequent days and have sufficient viral loads to infect susceptible animals.

he sensitivity analysis attempted to identify the most inluential parameters on the probability of truck contamination, particularly parameters whose values had been estimated on the basis of the experts' input. However, the outputs suggested that small incremental changes in the farm-level prevalence of

Figure 3: Distribution of posterior probabilities for the contamination of trucks with porcine reproductive and respiratory syndrome virus after application of one of the three diferent cleaning and disinfection protocols (w = washing; wd = washing and disinfection; and wdd = washing, disinfection, and drying), for a truck that was used by an infected finishing (fi) farm. Boxes represent inter quartile range of the distribution.

Figure 4: Risk plot showing sensitivity of the median probability that a truck will be infected with porcine reproductive and respiratory syndrome virus to changes in key model parameter values from those of the baseline model (1344): N ani (minimum number of infectious animals required to contaminate the truck), farm-level prevalence (F prev), animal-level prevalence (A prev) of the virus, and the probability of shedding animals (Shed ani) on the truck, respectively. Only representative sensitivity analysis scenarios are presented (supplementary material is available [Table S2] for median probabilities for other scenarios).

PRRS virus, in the animal-level prevalence of the virus, and in the percentage of shedding animals in the shipment, did not greatly affect the model outcome (median probability of truck contamination). The sensitivity analysis indicated that either decreasing the farm-level prevalence of the virus to 10% (from 50%) by participating in area regional control programs, or decreasing the animallevel prevalence of the virus to a similar level by adopting a number of PRRS elimination programs such as herd closure, all in-all out animal flow, and avoiding direct or indirect contacts between subpopulations within a farm, could decrease the probability for truck contamination by more than half. These two findings may have practical significance in controlling the spread of PRRS virus via shared transport.

Despite several simplifying assumptions, we believe the model has captured the underlying pathways leading to the contamination with PRRS virus of trucks used in the transportation of pigs on Canadian farms, from which infection can be transmitted to susceptible pigs. However, for some scenarios, only point estimates are presented for the probability of truck contamination, as the model could not produce uncertainties around these estimates due to the use of a step function in the model, which is a limitation of the model. Due to the lack of available data, the current model did not include pathways leading to eventual transfer of infection from such trucks to susceptible pigs or naive farms. However, the model could be further extended to elucidate such probabilities, as well as to estimate the indirect-contact transmission probability of spreading the PRRS virus via the sharing of trucks. A similar approach could be utilized in understanding the risk of truck sharing on the spread of other swine diseases where transportation has been implicated as a medium for viral spread, as appears to be the case for porcine epidemic diarrhea.²¹

Findings from this study have value to the Canadian swine industry in helping producers make informed decisions regarding the sharing of trucks among farms and guiding their selection of cleaning protocols for trucks. Given the current truck-sharing patterns among Canadian swine farms, where, for more than half of the shipments on any given day, the same truck has been used on more than one farm, 18 together with current biosecurity practices for truck cleaning in Canada, where only approximately one third of the trucks used for the shipment of pigs are cleaned after every shipment, ⁴⁰ the current model suggests that there is a sub-

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stantial risk for spread of PRRS virus through contaminated trucks. This risk could be largely eliminated either by properly washing, disinfecting, and drying trucks between shipments, by substantially decreasing the farmor animal-level prevalence of the PRRS virus, or by using designated trucks for each farm in an attempt to minimize sharing among farms. Planning shipments so that farms of similar PRRS status are visited in sequence (on the assumption that the PRRS virus status of each farm is known), or using dirty trucks for the transportation of market pigs and clean trucks for shipping gilts and young pigs to farms, may be strategies that minimize the transmission of PRRS virus via shared transportation. Cost is a major determinant for regular cleaning and disinfection of shipment trucks, so future studies to evaluate the cost and benefit of proper cleaning and disinfection of trucks should aid swine producers and transporters in making informed decisions.

Implications

- Findings from this study suggest that under current biosecurity practices, a substantial risk exists for the spread of PRRS virus due to truck sharing.
- Properly washing, disinfecting, and drying trucks between shipments could largely eliminate this risk.
- This model could also be utilized in understanding the risk of truck sharing on the spread of other swine diseases, such as porcine epidemic diarrhea.

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Conflict of interest

None reported.

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* Non-refereed reference.

Conversion tables

Weights and measures conversions

Temperature equivalents (approx)

Conversion chart, kg to lb (approx)

1 tonne = 1000 kg

1 ppm = 0.0001% = 1 mg/kg = 1 g/tonne

 1 ppm = 1 mg/L

Supplementary material

Modelling contamination of trucks used in the shipment of pigs infected with porcine reproductive and respiratory syndrome virus

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Data Description (Estimation of values/parameters)

Truck use

The proportion of trucks used (Tr.use_i) for two, three and four or more "j" farm visits on a given day and the proportion of trucks with more than the minimum number of shipped animals (Min_{ani}) for each of three "k" production types were obtained from the pilot pig traceability data.

Shipment size and travel time

he number of infectious animals in a given shipment, and the travel time were estimated as described below:

Prevalence of shedding animals on a truck

he prevalence of shedding animals (Shed. prev.k) was adjusted by the within-farm prevalence of PRRS (A.Prev.k). Expert judgement suggested that the proportion of animals shedding the virus (Shed.anim.k) varied according to the growth stage of the pigs being transported and it was suggested that 20%, 50% and 70% of weaned piglets, nursery pigs, and finishing pigs, respectively, would typically be shedding the virus. However, in doing so, we might have underestimated the risk by not accounting for those animals there were not seropositive yet but were still shedding the virus.

Several studies have reported very high within-farm prevalence of PRRS virus ranging from 80 -100% (Dee and Joo, 1994; Maes, 1997; Nodelijk et al., 2003). For this study, we used an animal-level prevalence (A.Prev.k) of 80% and evaluated the impact of this variable on the model outcome by carrying out sensitivity analysis.

Minimum number of shedding animals in a shipment

PRRS virus is excreted through urine, faeces and oral fluids of infected animals in addition to several other bodily secretions (Wills et al., 1997c; Bierk et al., 2001), though the dynamics of these shedding patterns is not well documented. Therefore, it was difficult to quantify the amount of PRRS virus likely to be present on any given shipment truck. We were interested in estimating the infectious potential of the trucks such that the virus could be transmitted to naive animals. We therefore assessed whether or not the trucks would likely have sufficient viral load to infect susceptible pigs. Dee and colleagues (2004b) demonstrated that the presence of two infectious pigs on a truck trailer for two hours was sufficient to transmit the virus to naive pigs on the subsequent introduction of these animals to the truck. Using these guidelines as a cut-of value, we categorized trucks as having sufficient infectious virus or not to transmit PRRS virus to naïve pigs (i.e. they must have transported at least two infectious animals and have had a travel time of at least two hours).

Minimum number of animals (Minani.k) on a truck to have at least two infectious animals

We used the hypergeometric distribution to estimate the minimum number of shedding animals needed in a shipment (Minani.k) for each production type, in order to have two infectious animals (Nani) on a truck that were shedding the virus. We used 95th percentile of shipment size (Shipsize.k), as recorded in the pilot pig traceability data, for each of the three production types as 'N', and the number of shedding animals, which was based on Shed.prev.k, as the 'm' parameter of the hypergeometric process. For psi, we used one, as the odds of drawing a shedding animal from the shipment was similar

to that of drawing an animal that was not shedding the virus. Psi is the odds of drawing shedding vs not shedding animal from the sample using hypergeometric distribution.

Shipment size (Shipsize) and Minimum shipment size (Minship.k)

The shipment size was simulated as a triangular distribution with 5th and 95th percentile of the shipment size recoded in the pilot pig traceability dataset for each of the three production types as minimum and maximum of the distribution. Based on this distribution for shipment size, the probability that a truck had minimum shipment size (Minship.k) or more animals than Minani.k was estimated using the step function available in OpenBUGS.

Infective Dose (Inf.dose)

Finally, we estimated the probability that a truck has an infective dose of PRRS virus (Inf.dose) if it shipped at least Minani and had a travel time (Travel) of at least two hours.

Additionally, since the infectious dose is related to the environmental conditions, we calculated the infective dose (Inf.dose) for warm and cold seasons. PRRS virus has been described as having a median infectious half-life of 14.6 hours (95% $CI = 12.6$ - 17.2) in pig manure at an ambient temperature of 220°C (Linhares et al., 2012). The PRRS virus has a comparatively longer half-life of 112.6 hours (95% CI = 103.2) - 123.8) in pig manure at an ambient temperature of 40°C (Linhares et al., 2012). We were guided by this information to extend the model to incorporate viral decay in order to quantify the risk that trucks would still be contaminated with PRRS virus on subsequent days in either warm and cold months.

For warmer months, we assumed that at least one infectious dose of virus would be present on trucks that have at least two infectious animals, that had been kept on the truck for at least two hours. So, based on viral decay with an assumed half-life of 15 hours, for at least one infectious dose of virus to be present during the 15 hours subsequent to the truck being used on Day 1, would require that at least four infectious animals (this is analogous to having two infectious doses of virus on Day 1) were present on the truck, and similarly for the truck to be infectious for the next 30 hours that at least eight infectious animals would need to have been present. On the other hand, for colder months we assumed that trucks with at least two infected animals could be considered to be infective for around five days, as the half-life of the virus is much longer (112.6 hrs at 40°C), and thus we did not attempt to quantify the

likelihood for time points beyond a one week duration from which the truck initially became infected.

Travel time

In order to estimate the amount of virus shed during transportation, the travel time was first estimated and then the probability that a given shipment was longer than two or more hours was computed. It was assumed that the most likely travel time in Ontario, Canada was around two hours, which corresponds to the travel time estimated by Dee et al. (2004b) for swine operations in Minnesota and was likely to vary between a minimum of half an hour and a maximum of six hours. We used this information to parameterise

a triangular (min, max) distribution in OpenBUGS. First, two similar uniform distributions were computed for travel time (min/2, max/2) and these distributions were summed together that yielded a triangular distribution. These equations provided a travel time distribution with mean and median of 3.25 hours.

The probability that a given shipment lasted for at least two hours was estimated using the step function to the distribution of travel time. The step function provided the probability of travel time equal to 1 if the travel time was more than two hours.

Table S1 continued on page 4

Table S1 continued: Summary posterior distribution of nodes and scenarios used in the Bayesian model simulated to evaluate the probability that a truck will be contaminated with PRRS virus at the end of a working day.

* 95% credible Interval

! Standard deviation

Table S2: Sensitivity analysis for the probability that a truck used by swine farms will be contaminated with PRRS virus at the end of a working day.

#CODES FOR THE MODEL

#A. Codes for OpenBugs

Model {

tr.use[1:3] ~dmulti(p.truse[], 100) p.truse[1:3]~ddirch(alpha[]) for(k in 1:3){ $alpha[k] < -1$ }

#Truck used by #tr.use2 ~ dbin(p.truse2, n) #tr.use3 ~ dbin(p.truse3, n) #tr.use4 ~ dbin(p.truse4, n)

#P that atleast one farm is infected $farm.pos2 \sim dbin(farm.prev, 2)$ F.inf.2<- step(farm.pos2-0.5)

 $farm.pos3 \sim dbin(farm.prev, 3)$ F.inf.3<- step(farm.pos3-0.5)

farm.pos $4 \sim$ dbin(farm.prev, 4) F.inf.4<- step(farm.pos4-0.5)

comb.prob <-F.inf.2*p.truse[1]+F.inf.3*p.truse[2]+F.inf.4*p. truse[3]

#P that the truck has more than Minimum infected animals

Nani.fa~dhyper(Minani.fa, m.fa, N.fa, 1) n1.fa~dunif(1,50) Minani.fa<-round(n1.fa) shipsize.fa1~dunif(5, 175) shipsize.fa2~dunif(5, 175) shipsize.fa<-(shipsize.fa1+shipsize.fa2)

Nani.fa1~dhyper(Minani.fa,m.fa, N.fa, 1) Nani.fa1.step<- step(Nani.fa1-2)

Minship.Fa <- step(shipsize.fa-Minani.fa)*Nani.fa1.step

Nani.nu~dhyper(Minani.nu, m.nu, N.nu, 1) n1.nu~dunif(1,30) Minani.nu<-round(n1.nu) shipsize.nu1~dunif(6, 350) shipsize.nu2~dunif(6, 350) shipsize.nu<-(shipsize.nu1+shipsize.nu2)

Nani.nu1~dhyper(Minani.nu,m.nu, N.nu, 1)

Nani.nu1.step<- step(Nani.nu1-2)

Minship.Nu <- step(shipsize.nu-Minani.nu)*Nani.nu1.step Nani.fi~dhyper(Minani.fi, m.fi, N.fi, 1) n1.fi~dunif(1,20) $Minani.fi <$ -round $(n1.fi)$ shipsize.fi1~dunif(3, 150) shipsize.fi2~dunif(3, 150) shipsize.fi<-(shipsize.fi1+shipsize.fi2)

Nani.fi1~dhyper(Minani.fi,m.fi, N.fi, 1) Nani.fi1.step<- step(Nani.fi1-2)

Minship.Fi <- step(shipsize.fi-Minani.fi)*Nani.fi1.step

#P that the travel time was more than two hours travel.time1~dunif(0.25, 3) travel.time2~dunif(0.25, 3)

travel.time<-(travel.time1+travel.time2)

##travel.time~dbeta(alpha.t, beta.t) #mean<-(min.t+lambda*mode.t+max.t)/(lambda+2) #alpha.t<-(mean-min.t)*(2*mode.t-min.t-max.t)/((mode.t mean)*(max.t-min.t)) #beta.t<-(alpha.t*(max.t-mean))/(mean-min.t) #lambda~dgamma(alpha1, beta1)

#travel.time ~ dnorm(mean, prec)C(0,) #mean <-(0.5+4*2+6)/6 #sd<-(6-0.5)/6 #prec<-1/pow(sd, 2) Travel <- step(travel.time-2)

#Evaluation of cleaning and disinfection protocols

#Truck_wash_protection node N.W.Prot ~ dbin(W.efficacy, N.W)

#Tr_washndisinfection_protection node N.Wd.Prot ~ dbin(Wd.efficacy, N.Wd)

#Tr_washndisinfectionndry_protection node N.Wdd.Prot ~ dbin(Wdd.efficacy, N.Wdd)

Prior distribns for efficacy - 50% W_c efficacy ~ dbeta $(0.5, 0.5)$ W d.efficacy ~ dbeta $(0.5, 0.5)$ Wdd.efficacy \sim dbeta(0.5,0.5)

#p.truse2 \sim dbeta(0.5,0.5) #p.truse3 \sim dbeta(0.5,0.5) #p.truse4 ~ dbeta(0.5,0.5)

#Prior for travel time #alpha1<-0.0001 #beta1<-0.0001

#Scenarios #Scenarios without cleaning S.2.fa <- F.inf.2*Minship.Fa*Travel S.2.nu <- F.inf.2*Minship.Nu*Travel S.2.fi <- F.inf.2*Minship.Fi*Travel S.3.fa<- F.inf.3*Minship.Fa*Travel S.3.nu <- F.inf.3*Minship.Nu*Travel S.3.fi<- F.inf.3*Minship.Fi*Travel S.4.fa <- F.inf.4*Minship.Fa*Travel S.4.nu <- F.inf.4*Minship.Nu*Travel S.4.fi <- F.inf.4*Minship.Fi*Travel S.fa <- comb.prob*Minship.Fa*Travel S.nu <- comb.prob*Minship.Nu*Travel S.fi <- comb.prob*Minship.Fi*Travel

#Scenarios with cleaning

S.fa.w <- (1-W.efficacy)*comb.prob*Minship.Fa*Travel S.fa.wd <- (1-Wd.efficacy)*comb.prob*Minship.Fa*Travel S.fa.wdd <- (1-Wdd.efficacy)*comb.prob*Minship.Fa*Travel S.nu.w <- (1-W.efficacy)*comb.prob*Minship.Nu*Travel S.nu.wd <- (1-Wd.efficacy)*comb.prob*Minship.Nu*Travel S.nu.wdd <- (1-Wdd.efficacy)*comb.prob*Minship.Nu*Travel S.fi.w <- (1-W.efficacy)*comb.prob*Minship.Fi*Travel S.fi.wd <- (1-Wd.efficacy)*comb.prob*Minship.Fi*Travel S.fi.wdd <- (1-Wdd.efficacy)*comb.prob*Minship.Fi*Travel }

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#B. Codes for R for combined outputs and for running the #Scenarios with cleaning CODA package "S.fa.w", "S.fa.wd", "S.fa.wdd", "S.nu.w", "S.nu.wd", #Truck risk Model "S.nu.wdd", library(R2OpenBUGS) "S.fi.w", "S.fi.wd", "S.fi.wdd" setwd("C:\\Users\\Risk_paper")) #par.c<-c("S.4.fi") #Data #Truck used by inits.2<- list(list(W.efficacy=0, Wd.efficacy=1, Wdd.efficacy=0.1), tr.use= c(49, 19, 32) #shipsize.fa=5, #travel time list(W.efficacy=0.05, Wd.efficacy=0.5, Wdd.efficacy=1), #shipsize.fa=50, N.W.Prot=0 N.W=20 list(W.efficacy=1, Wd.efficacy=0.5, Wdd.efficacy=0.1)) N.Wd.Prot=6 #shipsize.fa=100, N.Wd=10 tr.risk <- bugs(data.b, inits=inits.2 , parameters=par.b, N.Wdd.Prot=10 "Risk_model_openbugs_May19.txt", N.Wdd=10 Nani.fa=2 n.chains = 3, n.burnin=10000, n.iter = 50000, n.thin=1, Nani.nu=2 codaPkg=F, Nani.fi=2 working.directory = getwd(), clearWD=F, debug=F, #Now these parameters can easily be changed for the sensitivity DIC=F) analysis print(tr.risk, digits=4) farm.prev=0.5 #analysis convergence shed.prop.fa=0.2 library(coda) shed.prop.nu=0.5 tr.out=as.mcmc.list(tr.risk) shed.prop.fi=0.7 codamenu() 2 ani.prev=1 tr.out N.fa=351 #after this need to use interactive coda menu m.fa=round(ani.prev*shed.prop.fa*N.fa); m.fa gelman.plot(tr.out, bin.width = 10, max.bins = 50) N.nu=700 \bigcirc m.nu=round(ani.prev*shed.prop.nu*N.nu); m.nu $N.f = 250$ m.fi=round(ani.prev*shed.prop.fi*N.fi); m.fi data.b <- c("tr.use", "N.W.Prot", "N.W", "N.Wd", "N.Wd.Prot", "N.Wdd.Prot", "N.Wdd", "Nani.fa", "Nani.nu", "Nani.fi", "m.fa", "N.fa", "m.nu", "N.nu", "m.fi", "N.fi", "farm.prev") par.b <- c("farm.pos2", "farm.pos3", "farm.pos4", "comb.prob", "Minani.fa", "Minani.nu", "Minani.fi", "Minship.Fa", "Minship.Nu", "Minship.Fi", "Nani.fa1", "Nani.nu1", "Nani.fi1", "Nani.fa1.step", "Nannu1.step", "Nani.fi1.step", "travel.time", "Travel", "W.efficacy", "Wd.efficacy", "Wdd.efficacy", #Scenarios "S.2.fa", "S.2.nu", "S.2.fi", "S.3.fa", "S.3.nu", "S.3.fi", "S.4.fa", "S.4.nu", "S.4.fi", "S.fa", "S.nu", "S.fi",