



Contacts posing risks of disease introduction in swine breeding herds in Quebec, Canada: Is the frequency of contacts associated with biosecurity measures?

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ABSTRACT

The introduction of pathogens into swine breeding herds can occur through a variety of contacts involving people, animals, vehicle or various supplies. Appropriate biosecurity is critical to mitigate these risks. A retrospective study was conducted to describe contacts with swine breeding sites over a one-month period and to evaluate their association with biosecurity measures and site characteristics. As part of a larger project, sites which had a recent porcine reproductive and respiratory syndrome virus introduction were selected. A questionnaire, logbooks and pig traceability system were used for collecting data relative to persons or supplies entering the breeding unit, live pig transportation, service vehicles, other animal species, neighboring pig sites and manure spreading around the site. The 84 sites investigated had a median sow inventory of 675. A median of 4 farm staff and 2 visitors entered the breeding unit at least once over the one-month period. A total of 73 sites (87%) received visitor(s), mostly from maintenance and technical services. All sites received at least 3 supply deliveries (median of 8) including semen (99% of sites), small material and/or drugs (98% of sites), bags (87% of sites), and/or equipment (61% of sites). Live pig movements were observed in all sites, with a median number of 5 truck entries on the site or exits from the site. For feed mill, rendering and propane trucks, at least one entry was noted in $\geq 61\%$ of sites. For all service vehicle categories except feed mill and manure vacuum trucks, a single service provider was involved in each site. Dogs and cats were banned from all sites, but wild birds were observed in 8% of sites. Manure spreading within a 100 m radius of pig units was noted in 10% of the sites. With a few exceptions, biosecurity measures were not associated with the frequency of contacts. A 100-sow increase in sow inventory was associated with an increase of 0.34 in the cumulated number of staff entering the breeding unit, of 0.30 in the number of visitors and of 0.19 in the number of live pig movements. Live pig movements were also positively associated with vertically integrated farrow-to-wean (vs. independent farrow-to-wean) production and time interval of 4 weeks or more between farrowing (vs. less than 4). Considering the variety and frequency of contacts observed, biosecurity should be meticulously applied in all breeding herds to prevent endemic and exotic disease introduction.

1. Introduction

Numerous viral and bacterial pathogens responsible for exotic or

endemic diseases threaten the swine industry. African swine fever with its recent dissemination to several continents (Mason-D'Croz et al., 2020; You et al., 2021) is now the main exotic threat for the North

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American swine industry. Endemic diseases can also have a massive impact on the industry, for example the annual costs of porcine reproductive and respiratory syndrome (PRRS) are estimated at \$664 million in the USA (Holtkamp et al., 2013). For both exotic and endemic pathogens, application of biosecurity measures to address the different pathway(s) of transmission is of utmost importance to avoid their introduction in a herd or a country (Pitkin et al., 2009b; Alarcón et al., 2021).

For several diseases, the introduction of infected pigs or contaminated semen represents an important source of contamination (Le Potier et al., 1997; Amass and Clark, 1999). Indirect transmission pathways through fomites, such as clothes, supplies, feed or equipment also need to be considered for pathogens surviving in the environment such as porcine epidemic diarrhea (PED), African swine fever or PRRS viruses (Pitkin et al., 2009a; Kim et al., 2017; Kedkovid et al., 2020). Various types of vehicles can also enter the premises and can carry pathogens on their wheels over tens of kilometers (Dee et al., 2002) or within trailers having transported infected pigs, which supports their role as mechanical vectors (Lowe et al., 2014). Finally, aerosols can also be involved into disease transmission as shown for PRRS, foot-and-mouth disease, and *Mycoplasma hyopneumoniae* (Gloster et al., 1982; Otake et al., 2010).

Several biosecurity measures have been developed and promoted to mitigate the risk of introduction of pathogens into a herd. They include recommendations for people, animals, vehicles, semen, feed and other supplies, as well as for the general layout of the farm and location (Alarcón et al., 2021). While the application and effectiveness of specific biosecurity measures were often evaluated, the frequency and type of contacts experienced on farms have been much less described (Ribbens et al., 2009) which would be helpful in prioritizing biosecurity interventions. Even less evaluated was the association between on-farm biosecurity measures and contacts. The rationale is that the more contact the herd has, the more rigorous the biosecurity measures must be. The objectives of the current study were to 1) describe types and frequency of contacts in swine breeding sites over a one-month period prior to a new PRRSv introduction and 2) evaluate their association with specific biosecurity measures and site characteristics.

2. Material and methods

2.1. Study design and source population

As part of a larger project on PRRSv epidemiology, a retrospective observational study was conducted on swine breeding sites in Quebec, Canada. A swine breeding site was defined as a breeding unit or the collection of a breeding unit with one or more other pig units housing nursery or finishing stages, which belong to the same owner and are located at the same civic address; they could be farrowing, farrow-to-wean or farrow-to-finish operation and be part of a vertically integrated system (i.e., all pigs on the site belong to a large company incorporating multiple production sites, covering all stages of production from breeding to finishing, and providing most of the input such as feed, veterinary services and pig transportation) or be independent (i.e., otherwise). As part of a larger project conducted between August 1, 2014 and July 31, 2017 to assess sources of contamination, all swine breeding sites with ≤ 4 pig units, which had a recent PRRSv introduction during this period, and which were not in a selling process, were eligible for inclusion.

A recent PRRSv introduction was defined as the presence of PRRS clinical signs (fever, respiratory problems, abortions or other reproductive disorders) according to the herd veterinarian along with the detection of an ORF5 PRRSv sequence which met one of the following criteria: 1) the sequence had $\geq 97.5\%$ genetic similarity with a reference vaccine strain in a non-vaccinated herd; 2) the sequence had $\leq 92\%$ genetic similarity with sequences previously detected in the herd over the last four years; 3) the sequence had between 92% and 97.5% genetic similarity with a sequence previously detected in the herd over the last

four years, if this previous detection was followed by a successful PRRSv eradication confirmed by the veterinarian.

In the absence of sequences submitted in the last four years to compare with, the veterinarian had to confirm that the herd was PRRSv negative prior to clinical signs; if so it was an eligible case. This detection process was based on weekly analyses of the Laboratoire d'épidémiologie et de médecine porcine database (LEMP-DB; Swine Medicine and Epidemiology Laboratory), that gathers all PRRSv sequences from sequencing activities performed in Quebec since 2010 following data sharing agreements between veterinarians, diagnostic laboratories and LEMP (Lambert et al., 2019). The date of PRRSv introduction was estimated as the date of onset of the first clinical signs compatible with the PRRS episode associated with the submission. To be eligible, the sequencing had to be performed within 30 days from the first clinical signs to decrease potential recall bias.

All eligible sites were recruited following the owner's informed consent, and thus no sample size was calculated. When a site experienced more than one recent PRRSv introduction as defined above over the study period, only the first introduction was considered for inclusion. Reasons for refusal or nonparticipation were recorded to assess potential non-response bias. Participants were given a CAD 200 compensation following completion of the interview.

2.2. Data collection

Data on the general characteristics of the breeding sites and on contacts at risk for pathogen introduction in a herd were collected. Six types of risks were covered: 1) persons entering the breeding unit, 2) supplies entering the breeding unit, 3) live pig movements, 4) service vehicles entering the premises, 5) other animal species in breeding unit, and 6) neighboring pig sites and manure spreading around breeding site. For each type of risk, contacts were evaluated over the one-month period prior to the estimated date of the PRRSv introduction on the breeding site. This period allowed to capture a maximum of types of contacts occurring at different frequencies for the current study, but also to list most of the events that could have occurred during the incubation period of the disease to identify potential sources of contamination, which was one of the objectives of the larger project. The frequency of contacts (e.g., number of visitor entries), the number of sources of contacts (e.g., number of individual persons entering the site at least once) and of specific biosecurity measures in place to mitigate the risks associated with these contacts were collected (see details below). The compliance to the reported biosecurity measures over the one-month period was also assessed semi-quantitatively (always, occasionally, or never) by producers.

A questionnaire designed by the research team with mainly semi-closed questions. The relevance and clarity of the questions were assessed by a swine veterinarian. The first sampled herds were used for pre-validation. When an ambiguous question was noted, the questionnaire was revised before being used in subsequent herds, while previous participants were recontacted to verify their answers to the updated question. The questionnaire was written in French for the benefit of the participants and is available upon request. The questionnaire was completed during a 2 hr in-person interview with the owner of the site for independent sites or sites under contract in a vertically integrated system, or with the employee responsible for sites owned by a vertically integrated system. A single person of the research team performed all interviews. All data were collected through this questionnaire except for complementary information obtained on persons entering the breeding unit from farm logbooks (when available), on pig movements from the Quebec provincial traceability system (Les Éleveurs de porcs du Québec, 2021) and on neighboring pig sites from the Veille sanitaire provinciale (VSP; Provincial sanitary monitoring database).

2.2.1. Persons entering the breeding unit

The name of each farm staff or visitors that accessed the inside of the

breeding unit during the one-month period was noted. Farm staff was defined as full-time or part-time employees, pig owners, or site owners. Visitors were classified as maintenance personnel (e.g., maintenance of plumbing, electricity, welding, building, pig watering system, swine manure storage system, feeding system, propane tanks, fuel tanks), technical service personnel (animal health technician, agronomist, company representative, information technology/computer representative, laundry), exterminator, washing unit worker, teaching/training/research specialist, veterinarian, inspector (inspection or certification by the Canadian Pork Excellence, Canadian Food Inspection Agency or insurance companies), or any others. For each visitor, the number of visits was noted and defined as the number of days with at least one entry in the breeding unit over the one-month period. The number of visits was derived from farm logbooks if available; otherwise, they were obtained from the questionnaire using a categorical scale (1, 2, 3, ≥ 4 visits).

The following biosecurity measures related to people entrance protocol in the breeding unit during the one month were assessed separately for all farm staff combined and for all visitors combined: locking of main entrance door, signature of logbook, and shower-in, or a delimited entrance with change of boots, change of coveralls, hand washing with soap or disinfectant or wearing of gloves. A delimited entrance was defined as the presence of a visual (line) or physical (bench, raised floor) separation between the contaminated and clean areas of the entrance.

2.2.2. Supplies entering the breeding unit

Supplies entering the breeding unit over the one-month period were categorized into four categories: 1) small material (needles, swabs, gloves, acidifiers, disinfectants, letters, camera) and/or drugs, 2) equipment (repair or maintenance equipment, building material, toolbox or other specialized equipment as hog catchers or stunning pistol), 3) semen, and 4) bags (starter feed, minerals, drying agent for piglets, milk substitute). For each category, the number of deliveries (1, 2, 3, ≥ 4) and the number of supply distributors over the one-month period were noted. For semen deliveries, a question on its origin (i.e., same site, different site from the same company, approved semen collection centre, other external site) was added. A delivery was defined as one or more entry in the breeding unit of the same category of supply coming from the same supply distributor on the same day.

The way of delivering for each type of supply was noted: picked up by the owner or delivered outside the controlled access zone (CAZ), delivered within the CAZ but in a building separated from the breeding unit, or delivered within the breeding unit. The CAZ is the restricted site perimeter delimited by a gate or indicated by a sign and designed to limit the entrance of unauthorized vehicles and prevent site contamination. Next, the method used to receive the supply at the entrance of the breeding unit was noted (i.e., external portal, pass-through window, specific delivery reception room within the breeding unit, or absence of a physically delimited entry lock). The sanitization protocol (washed with soap, and/or disinfected, or fumigated, or none) was recorded for all types of supply except semen.

2.2.3. Live pig movements

Information on live pig entering or exiting the site within the one-month period was extracted from the Quebec provincial traceability system (*Les Éleveurs de porcs du Québec*, 2021). For each movement of a truck transporting pigs, data were obtained on the date, direction of pig movement (entry or exit), pig category (gilts, weaned piglets ≈ 5 kg, growing pigs ≈ 25 kg, finishing pigs, or culled sows), and the trailer licence plate number. For entry of gilts, the animal source type (self-replacement on the site, self-replacement from another site of the same company, external supplier) was recorded. For each pig category, a movement was defined as an entry or an exit on the site of a truck transporting pigs. Thus, if the same truck made two trips on the same day to transport pigs from one source to the site, it was recorded as two movements.

For each pig category with reported movement, the following biosecurity measures were recorded: vehicle washing before entering the premises (for exit movements), back-to-back strategy to pick up pigs, washing and disinfection of the loading dock after each use, and access of the delivery man within the pig unit. For gilt entry, the presence of a quarantine period was also documented as well as the pig flow (all-in-all-out, continuous) and the washing and disinfection of facilities applied between each group of gilts.

2.2.4. Service vehicles entering the premises

For each category of service vehicle entering the premises (feed mill, fuel tanker, propane, rendering, manure vacuum, waste collection, winter services), the number of vehicle entry (1, 2, 3, ≥ 4) and the name of the company were obtained from the interview. A vehicle entry was defined as one or multiple entry on the site of the same category of vehicle of the same company on the same day.

Regarding biosecurity practices, for feed mill, rendering and manure vacuum trucks, information on the usual entrance path on the site (i.e., same or different from the main entrance of the site used by farm staff) was asked. For manure vacuum trucks, additional data were collected: use of equipment only for this site and washing of equipment before the entry on the site. In addition, when rendering services were used for dead pig disposal, the presence of following biosecurity measures was gathered: carcasses picked up at ≥ 100 m from a pig unit of the site, outside or inside (freezer) rendering bin and the re-entrance protocol of staff accessing the rendering bin (i.e., changing boots, coveralls and washing hands).

2.2.5. Other animal species in breeding unit

The presence of dogs, cats, or wild birds in the breeding unit during the one-month period were documented using the questionnaire. The presence or absence of a wire mesh bird screen in the breeding unit was noted for each breeding unit.

2.2.6. Neighboring pig sites and manure spreading around breeding site

The number of pig sites in the area surrounding each breeding site was estimated. Data on all pig sites in Quebec registered in the VSP were obtained in September 2016. Each site was georeferenced using the approximated centroid of all pig units. Geographic coordinates were projected to Lambert conic conform coordinates and Euclidian distances between each pair of sites were calculated in ArcGIS (version 10.6; Esri, Redlands, CA). The number of sites within a 1, 2, 3, 4 and 5 km radius of each study site was calculated. The presence of manure spreading at less than 100 m from at least one pig unit of the site during the one-month period, and the source of manure (from the site vs. external site) were documented using the questionnaire. The presence or absence of an air filtration system was noted for each site.

2.3. Statistical analyses

All data management and statistical analyses were performed using SAS software (version 9.4; SAS Institute, Cary, NC). Descriptive statistics were used to present the general characteristics of the breeding sites. For each type of contact, the number of sites having experienced the contact at least once in the one-month period was described, and then the frequency of contacts was described for these sites. The distribution of different sources of contacts (i.e., the number of persons, supply distributors, pig transportation vehicles and service providers having at least one contact with the site over the one-month period) was also described.

For each type of contact, the associations between the frequency of contacts and related biosecurity measures were assessed among sites reporting this specific contact. For staff and visitors, the number of persons was used instead of the number of contacts considering the absence of data collection on the frequency of staff visits. The frequency of contacts (or persons) was dichotomized using the respective median

(among sites reporting the contact). Biosecurity measures were also dichotomized as “always” and “occasionally and never” applied, this latter being a merge of the two original categories. The associations were evaluated using exact chi-square tests. An alpha value of 0.05 was used to determine statistical significance. For contingency tables larger than 2×2 , post-hoc pairwise comparisons were done with Bonferroni adjustment.

The associations between the number of staff, number of visitors and number of live pig movements as outcomes and characteristics of the sites were evaluated. These outcomes were selected as the information was collected as continuous variables and considering their importance for disease transmission. The sow inventory, type of production (farrow-to-wean, farrow-to-grow, farrow-to-finish), type of production system (independent, vertically integrated), time interval between batch farrowing (<4 vs. ≥ 4 weeks) and number of pig sites in a 5 km radius were considered as explanatory variables. The correlation between each pair of these variables was first evaluated. As the type of production and production system variables were highly correlated, they were merged prior to modelling. Full multivariable linear regression models were built including all explanatory variables. A backward selection strategy was then used to select parsimonious final models. Variables with $P > 0.05$ were sequentially removed, except if their removal was associated with a $> 30\%$ change in coefficient estimates of significant predictors in the model, which was suggestive of confounding. Assumptions of normality, linearity and homoscedasticity of residuals were assessed using histograms, scatter plots of residuals against predicted values and scatter plots of residuals against each predictor.

3. Results

3.1. Study population

A total of 105 recent PRRSv introductions were detected in 96 breeding sites between August 1, 2014, and July 31, 2017. Site owner's informed consents were obtained for the first PRRSv introduction from 84 (88%) of the 96 breeding sites. Reasons for refusal were lack of interest or time.

3.2. General characteristics of breeding sites

The 84 swine breeding sites included in our study were mainly (62/84, 74%) farrow-to-wean (Table 1). Approximately half of the sites (48%) were independent and the others (52%) were part of one of eight large vertically integrated swine production systems. A majority of sites

Table 1

General characteristics of the 84 swine breeding sites included in the study in Quebec, Canada, 2014–17.

General characteristics	No.	%
Type of production		
Farrow-to-wean	62	74
Farrow-to-grow	3	3
Farrow-to-finish	19	23
Type of production system		
Independent	40	48
Vertically integrated	44	52
Time interval between batch farrowing		
< 4 weeks	35	42
≥ 4 weeks	49	58
Number of pig units on site		
1 unit	61	73
2 units	16	19
3 or 4 units	7	8
Administrative region		
Montérégie	40	48
Chaudière-Appalaches	30	36
Centre-du-Québec	8	9
Others	6	7

(73%) had only one pig unit, and 84% were located in two administrative regions, Montérégie or Chaudière-Appalaches. The median (Q1-Q3) was 675 (328–1020) for number of sows and 1046 (693–1523) for total pig inventory on the sites.

3.3. Description of contacts

3.3.1. Persons entering the breeding unit

The median (Q1-Q3) number of farm staff individuals that entered the breeding unit over the one-month period was 4 (3–6) and the maximum was 16. Eighty-seven percent of the sites had at least one visitor entering the breeding unit (Table 2). The median (Q1-Q3) and maximum number of individual visitors were 2 (1–5) and 13 among the 84 sites, respectively. A majority of sites (63%) were visited at least once for technical services over the one-month period (Table 2). A single person was involved in these visits for the category of exterminator, veterinarian, or inspector, whereas multiple individuals were often noted for other categories of visitors, especially for “maintenance” as well as “teaching, training and research” categories.

3.3.2. Supplies entering the breeding unit

All sites had at least 3 supply deliveries entering the breeding unit over the one-month period, from at least 2 supply distributors (Table 3). The median number of supply deliveries per site was 8. Small material and/or drugs were delivered at least once in 98% of the 84 breeding units, with 58 sites having received at least 2 deliveries and 35 sites having received this material from 2 or 3 supply distributors. Equipment was delivered in 61% of the sites, for either regular maintenance, major repairs, or construction of new buildings. Semen was delivered in all sites, with the exception of one start-up site. The number of semen deliveries was ≥ 4 in most (95%) of the remaining 83 sites. Semen originated from the same commercial insemination centre in 73% of sites and from boar studs of the vertically integrated production system in 27% of sites. Most breeding sites (87%) received at least one delivery of bags (e.g., starter feed for piglets, minerals).

3.3.3. Live pig movements

Pig movements were observed in all sites over the one-month period and involved at least 2 vehicles in 86% of the sites (Table 4). The median (Q1-Q3) and maximum number of pig movements per site were 5 (3–8) and 16, respectively. Movements of gilts, weaned piglets and culled sows were reported in a majority of sites (62%, 75% and 89%, respectively). Replacement gilts were provided by a genetic company (40%), a vertically integrated system (58%), or self-replacement (2%).

3.3.4. Service vehicles entering the premises

Almost all (98%) breeding sites had at least one entry of a service vehicle over the one-month period (Table 5). With the exception of feed mill and manure vacuum trucks, all vehicles from the same category entering a site were from the same company.

Feed mill truck entered in 93% of breeding sites. Most of these sites received ≥ 4 feed deliveries and most received feed from a single provider. The vast majority (96%) of vehicles used for feed deliveries were from feed companies or large production system.

Three quarters (74%) of the sites had at least one entry of a pig carcass rendering truck during the one-month period. All renderings were performed by one of the 3 rendering companies identified or by a truck from a large production system which had its own transport of pig carcasses to rendering services.

Manure vacuum trucks were observed in 36% of the breeding sites. Vehicles used to empty the manure pit either belonged to a commercial company (40%), to a large production system (40%) or were privately owned (20%).

3.3.5. Other animal species in breeding unit

Dogs and cats were banned from all sites. Wild birds were observed

Table 2

Number (%) of sites with different categories of visitors entering the breeding unit over a one-month period and their distribution (number) according to the cumulative number of visits and of visitors in 84 swine breeding sites in Quebec, Canada, 2014–17.

Categories of visitors	No. (%) of visited sites	Distribution among visited sites							
		No. of visits				No. of visitors			
		1	2	3	≥ 4	1	2–4	5–9	≥ 10
Maintenance	37 (44)	10	5	2	20	11	23	3	0
Technical services	53 (63)	17	16	8	12	44	9	0	0
Exterminator	35 (42)	34	1	0	0	35	0	0	0
Washing unit worker	9 (11)	2	1	2	4	7	2	0	0
Teaching, training, research	10 (12)	4	2	0	4	4	4	2	0
Veterinarian	21 (25)	19	2	0	0	21	0	0	0
Inspector	6 (7)	6	0	0	0	6	0	0	0
Others	6 (7)	3	2	0	1	4	2	0	0
Any category	73 (87)	16	9	3	45	21	31	16	5

Table 3

Number (%) of sites with different categories of supply entering the breeding unit over a one-month period and their distribution (number) according to the cumulative number of deliveries and of supply distributors in 84 swine breeding sites in Quebec, Canada, 2014–17.

Categories of supply	No. (%) of sites with deliveries	Distribution among sites reporting deliveries							
		No. of deliveries				No. of supply distributors			
		1	2	3	≥ 4	1	2	3	≥ 4
Small material and/or drugs	82 (98)	24	29	7	22	47	30	5	0
Equipment	51 (61)	33	6	2	10	40	5	4	2
Semen	83 (99)	1	3	0	79	78	5	0	0
Bag	73 (87)	55	12	2	4	67	5	1	0
Any category	84 (100)	0	0	1	83	0	13	25	46

in breeding unit in 7 of the 84 sites (8%).

3.3.6. Neighboring pig sites and manure spreading around the breeding site

The proportion of sites with ≥ 1 neighboring pig sites increased from 56% within a 1 km radius up to 88% in a 5 km radius (Table 6).

Manure spreading within a 100 m radius of at least one pig unit was recorded in 8 of the 84 sites (10%). The source of manure was either from the same pig site (62%), or from an external one (38%).

3.4. Associations between contacts and biosecurity measures

3.4.1. Persons entering the breeding unit

No association was observed between the entry of ≥ 4 staff on the breeding unit over the one-month period and the biosecurity measure

Table 4

Number (%) of sites with different categories of pig movements over a one-month period and their distribution (number) according to the cumulative number of movements and of individual vehicles in 84 breeding sites in Quebec, Canada, 2014–17.

Categories of movements and pigs	No. (%) of sites with pig movements	Distribution of sites reporting pig movements							
		No. of movements				No. of vehicles ¹			
		1	2	3	≥ 4	1	2	3	≥ 4
Reception (entry) of pigs									
Gilts	52 (62)	33	15	4	0	44	6	0	0
Expedition (exit) of pigs									
Weaned piglets ≈ 5 kg	63 (75)	11	27	7	18	38	20	3	0
Growing pigs ≈ 25 kg	6 (7)	1	3	1	1	5	0	0	0
Finishing pigs	16 (19)	1	1	4	10	7	6	3	0
Culled sows	75 (89)	46	17	7	5	58	15	0	0
Any category	84 (100)	1	8	13	62	10	27	32	12

¹ Eight pig movements distributed among the various pig categories and involving 3 sites had missing data on vehicle licence plate number. These 3 sites were excluded from the number of vehicles distribution.

investigated, i.e., locking of the main entrance door, signature of logbook and entrance protocol (Table 7). However, sites with ≥ 3 visitors had a higher probability of always using a signed logbook for visitors and of applying a specific entrance protocol for all visitor entries.

3.4.2. Supplies entering the breeding unit

The three biosecurity measures related to supply entry in the breeding unit (i.e., delivery outside the breeding unit, reception method within breeding unit and sanitization) were not associated with the frequency of each type of supply delivery (Table 8).

3.4.3. Live pig movements

For expedition of growing pigs ≈ 25 kg, no association with biosecurity was tested due to the limited number of sites (n = 6; Table 4) reporting this movement. The probability of using either a back-to-back or washing and disinfection of the loading dock and vehicle before entrance on the site was not associated with the frequency of movements, except for reception of gilts (Table 9). Sites reporting only one gilt entry over the one-month period had a higher probability of applying this biosecurity measure compared to sites reporting more than one entry. No association was found between delivery man access within pig unit and frequency of movements, regardless of the category of pig movement (Table 9).

Among breeding herds that had received gilts during the one-month period, quarantine was located off-site (n = 3), on-site in a separated unit (n = 5), within the breeding unit (n = 11) or absent (n = 33). A quarantine was defined as optimal if it was in an all-in-all-out pig flow with washing and disinfection between batches, and as suboptimal otherwise, when present. No association was observed between the quarantine protocol for gilts and the frequency of gilt reception on the site. Among the 19 sites reporting more than one reception of gilts, the quarantine was optimal in 4 (21%) sites, suboptimal in 3 (16%), and absent in 12 (63%). These proportions were similar among the 33 sites

Table 5

Number (%) of sites with entry of different categories of service vehicle over a one-month period and their distribution (number) according to the cumulative number of entries and of delivery companies in the 84 breeding sites in Quebec, Canada, 2014–17.

Categories of service vehicleNo. (%) of sites with entry	Distribution in sites reporting entry of vehicles								
	No. of entries				No. of companies				
	1	2	3	≥ 4	1	2	3	≥ 4	
Feed mill truck (93)	78	5	6	3	64	70	6	2	0
Fuel tanker truck (21)	18	15	2	0	1	18	0	0	0
Propane truck (61)	51	34	10	2	5	51	0	0	0
Rendering truck (74)	62	20	24	1	17	62	0	0	0
Manure vacuum truck (36)	30	16	6	1	7	28	2	0	0
Waste collection vehicle (49)	41	37	2	2	0	41	0	0	0
Winter service vehicle (18)	15	14	1	0	0	15	0	0	0
Any category (98)	82	1	3	0	78	3	8	27	44

Table 6

Number (%) of sites with at least one neighboring pig site and their distribution (number) according to the number of neighboring pig sites for different radii in 84 breeding sites in Quebec, Canada, 2014–17.

Neighborhood radius	No. of sites (%) with ≥ 1 neighboring pig site	Number of neighboring pig sites			
		1	2–4	5–9	≥ 10
1 km	47 (56)	22	23	1	1
2 km	65 (77)	14	29	17	5
3 km	71 (85)	4	20	26	21
4 km	72 (86)	3	8	18	43
5 km	74 (88)	3	3	11	57

Table 7

Number (%) of sites reporting biosecurity measures always applied at the entrance of the breeding unit for staff or visitors, according to the number of staff or visitors entering the breeding unit over a one-month period in Quebec, Canada, 2014–17.

Category of persons	No. of sites	No. (%) with the main entrance door locked	No. (%) with a signed logbook	No. (%) with an entrance protocol ¹
Staff (n = 84 sites)				
< 4 staff	35	19 (54) ^a	1 (2.9) ^a	18 (51) ^a
≥ 4 staff	49	30 (61) ^a	5 (10) ^a	31 (63) ^a
Visitors (n = 73 sites)				
< 3 visitors	33	18 (54) ^a	7 (21) ^a	19 (58) ^a
≥ 3 visitors	40	30 (75) ^a	21 (53) ^b	32 (80) ^b

^{a,b} For each biosecurity measure and type of person combinations, the proportions with different superscripts are significantly different ($P < 0.05$) based on exact chi-square tests.

¹ Shower-in or delimited entrance with changing coveralls, changing boots, and washing hands or wearing gloves.

reporting only one reception of gilts; the quarantine was optimal in 6 (18%) sites, suboptimal in 6 (18%), and absent in 21 (64%) sites.

3.4.4. Service vehicles entering the premises

The access of the site by a different entrance path from the staff main entrance was not associated with the frequency of site entries by feed mill, manure vacuum or rendering trucks (Table 10). Similarly, for

Table 8

Number (%) of sites reporting biosecurity measures always applied for supply deliveries according to the frequency of different categories of supply entering the breeding unit over a one-month period in Quebec, Canada, 2014–17.

Categories of supply	No. of sites	No. (%) with delivery outside the breeding unit ¹	No. (%) with reception protocol within breeding unit ²	No. (%) with sanitization of supply
Small material and/or drugs (n = 82 sites)				
≥ 2 deliveries	58	25(43) ^a	14 (24) ^a	22 (38) ^a
1 delivery	24	12 (50) ^a	6 (25) ^a	11 (46) ^a
Equipment (n = 51 sites)				
≥ 2 deliveries	18	1 (5.6) ^a	2 (11) ^a	8 (44) ^a
1 delivery	33	9 (27) ^a	0 (0) ^a	14 (42) ^a
Semen (n = 83 sites)				
≥ 4 deliveries	79	35 (44) ^a	38 (48) ^a	-
1–3 deliveries	4	1 (25) ^a	2 (50) ^a	-
Bag (n = 73 sites)				
≥ 2 deliveries	18	13 (72) ^a	3 (17) ^a	4 (22) ^a
1 delivery	55	24 (44) ^a	15 (27) ^a	19 (35) ^a

^{a,b} For each biosecurity measure and category of delivery combinations, the proportions with different superscripts are significantly different ($P < 0.05$) based on exact chi-square tests.

¹ Pick-up by the owner, delivery outside the controlled access zone (CAZ), or within the CAZ but in a building separated from the breeding unit.

² Delivery using an external portal, a specific delivery reception room within the breeding unit or a pass-through window at the breeding unit entrance.

Table 9

Number (%) of sites reporting biosecurity measures always applied for live pig movement according to the frequency of different categories of pig movements in breeding sites over a one-month period in Quebec, Canada, 2014–17.

Categories of pig movements	No. of sites	No. (%) with back-to-back ² or washing and disinfection of the loading dock and vehicle ³ before entrance on the site	No. (%) with delivery man never allowed within pig unit
Reception of gilts (n = 52 sites)			
> 1 movement	19	0 (0) ^a	18 (95) ^a
1 movement	33	8 (24) ^b	31 (94) ^a
Expedition of 5 kg piglets (n = 63 sites)			
> 2 movements	25	11 (44) ^a	24 (96) ^a
≤ 2 movements	38	19 (50) ^a	34 (89) ^a
Finishing pigs (n = 16 sites)			
> 4 movements	8	3 (38) ^a	7 (88) ^a
≤ 4 movements	8	0 (0) ^a	7 (88) ^a
Expedition of culled sows (n = 74 sites) ¹			
> 1 movement	28	8 (29) ^a	27 (96) ^a
1 movement	46	14 (30) ^a	43 (94) ^a

^{a,b} For each biosecurity measure and pig category combinations, the proportions with different superscripts are significantly different ($P < 0.05$) based on exact chi-square tests.

¹ Of the 75 sites that reported culled sow movement, one had missing data on biosecurity and was excluded.

² Intermediate vehicle to transfer pigs, avoiding direct contact between external vehicle and pig units. Back-to-back was always used in 8 sites for gilts reception and in 1 and 14 sites for the expedition of 5 kg piglets and culled sows, respectively

³ Except for gilts.

rendering trucks, no association was observed between the frequency of entry and carcass picking up at ≥ 100 m from a pig unit (Table 10).

No association was found between staff access to rendering bin and frequency of rendering truck entrance on the site. Among the 17 sites reporting ≥ 4 entries of a rendering truck, 1 (6%) had a freezer located in the breeding unit, 10 (59%) had an outside rendering bin with a re-entrance protocol (changing boots, coveralls and washing hands), and 6 (35%) had an outside rendering bin without a re-entrance protocol always implemented. Among the 45 sites reporting < 4 entries of a rendering truck, these numbers were 13 (29%), 13 (29%) and 19 (42%), respectively.

Regarding equipment used by manure vacuum truck, 3 sites used it on the site only, 6 sites used it on other pig sites but was washed before site entrance, and 21 had equipment used on other pig sites which was not always washed. An overall association ($P < 0.01$) was observed between the type of equipment used and frequency of manure vacuum trucks. Based on post-hoc comparisons, among the 27 sites having shared equipment, the probability of using equipment washed before site entrance (vs. not always washed) was higher ($P < 0.01$) in sites with more than one vacuum truck entries (6/12, 50%) compared to sites with only one vacuum truck entry (0/15, 0%).

3.4.5. Other animal species in breeding unit

Most breeding sites (83%) had wire mesh bird screens in air inlets. Only 1% of the 70 sites with wire mesh bird screen in air inlets reported the presence of wild birds in pig units compared to 43% of the 14 sites without these screens ($P < 0.001$).

3.4.6. Neighboring pig sites and manure spreading around the breeding site

Of the 84 breeding sites, only 4 (5%) had an air filtration system, and of these, only 2 were in function during the one-month period of our study. Considering this data limitation, no association was tested between air filtration and the number of neighboring pig sites or manure spreading around the site.

3.5. Associations between contacts and general characteristics of breeding sites

A 100-sow increase was associated with an increase of 0.34 in the number of staff entering the breeding unit over the one-month period, of 0.30 in the number of visitors, and of 0.19 in the number of live pig movement (Table 11). A higher number of live pig movements was also observed on farrow-to-wean vertically integrated sites compared to others and on sites with < 4 -week interval between batch farrowing compared to ≥ 4 -week interval. No departure from models' assumption was noted.

Table 10

Number (%) of sites reporting biosecurity measures always applied for entry of service vehicles according to the frequency of entries of different service vehicles on the breeding site over a one-month period in Quebec, Canada, 2014–17.

Categories of service vehicle	No. of sites	No. (%) with access to site by a different entrance path from the staff main entrance	No. (%) with carcasses picked up at ≥ 100 m from a pig unit of the site
Feed mill trucks (n = 78 sites)			
≥ 4 entries	64	4 (6.3) ^a	-
1–3 entries	14	0 (0) ^a	-
Manure vacuum trucks (n = 30 sites)			
≥ 2 entries	14	0 (0) ^a	-
1 entry	16	3 (19) ^a	-
Rendering trucks (n = 62 sites)			
≥ 4 entries	17	6 (35) ^a	15 (88) ^a
1–3 entries	45	21 (47) ^a	28 (62) ^a

Table 11

Parameter estimates with 95% confidence intervals (CI) of final multivariable linear regressions modelling the total number of staff, total number of visitors and total number of live pig movements over a one-month period as outcomes according to site characteristics in 84 swine breeding sites in Quebec, Canada, 2014–17.

Model outcomes and characteristics of breeding sites	No. of sites	Parameter estimates		
		b	95% CI	P-value
Outcome = Total number of staff (model P-value (F test) < 0.001 and $r^2 = 0.37$)				
Sow inventory (for each 100-sow increase)	84	0.34	0.25–0.44	< 0.001
Outcome = Total number of visitors (model P-value (F test) < 0.001 and $r^2 = 0.30$)				
Sow inventory (for each 100-sow increase)	84	0.30	0.20–0.40	< 0.001
Outcome = Total number of live pig movements (model P-value (F test) < 0.001 and $r^2 = 0.42$)				
Sow inventory (for each 100-sow increase)	84	0.19	0.04–0.34	0.01
Production type and system				
Farrow-to-grow or farrow-to-finish*	22	1.63	-0.25–3.50	0.09
Farrow-to-wean, vertically integrated	42	1.82	0.22–3.43	0.03
Farrow-to-wean, independent	20	Ref		
Time interval between batch farrowing				
< 4 weeks	35	3.72	2.40–5.03	< 0.001
≥ 4 weeks	49	Ref		

* With the exception of 2 vertically integrated farrow-to-finish sites, all sites were independent.

4. Discussion

Our study provides new insights on the types and frequencies of contacts that could pose a risk of pathogen introduction on swine breeding sites in Quebec, Canada. The voluntary participation of 87% of eligible sites was considered high. The average sow inventory of 675 in our participating breeding sites was higher than the average sow inventory of 458 in Quebec breeding sites (VSP; Provincial sanitary monitoring database). In addition, our source population was limited to swine breeding sites with a recent PRRSv introduction. This may question the validity of generalizing our results to all breeding herds in Quebec, as larger herds or herds that had not experienced a new PRRSv introduction might have had different biosecurity measures or types and numbers of contacts. However, given that at that period (2014–2017), 75% of all swine breeding sites in Quebec were considered positive to PRRSv, i.e., previously exposed to a PRRSv strain (CDPQ, 2016), we believe that our study population is likely representative of the majority of large swine breeding herds in Quebec. Of note, PRRS shows a seasonal pattern in certain regions, being more frequent during the cold months. Although we do not suspect seasonal differences in the frequency of most contacts considering the year-round production, the frequency of some contacts might have been underestimated as compared to the general herd population, for example manure spreading or else, over-estimated for entry of winter service vehicle. With regards to PRRSv, since the association between contacts and virus introduction was not quantified in this project, the results should only be used to generate hypotheses on potential transmission pathways.

A considerable traffic of staff and visitors was observed with a median of 4 different staff and 2 different visitors entering the breeding unit of the site at least once over a one-month period. These numbers reached up to 16 staff and 13 visitors. Given that fomites carried by people can act as mechanical vectors for many pathogens (Fedorka-Cray et al., 1997; Otake et al., 2002; Pitkin et al., 2009a), an entrance protocol needs to be implemented to mitigate the risk of pathogen introduction. A 3-zone delimited entrance with changing coveralls and boots and washing hands for visitors, with an intermediate area establishing a clear delimitation between clean (inside) and dirty (outside), can be

easily set up. Both shower and 3-zone entrance were shown to be effective in reducing the mechanical transmission of some pathogens (Pileri and Mateu, 2016; Alarcón et al., 2021). In addition, keeping the entrance door locked serves both to avoid any unexpected visitor and to increase compliance to the entrance protocol. In fact, visitors unfamiliar with the entrance protocol should be closely monitored to reduce risks. This could be particularly relevant for some type of visitors such as maintenance people, considering the relatively high frequency of these visits along with the diversity of people conducting them. Finally, keeping a signed logbook for all entries is simple but crucial, especially during outbreaks when traceback investigations are set up to control emerging or exotic diseases. Fortunately, breeding sites with a higher number of visitors also had a higher frequency of shower or delimited entrance and of signed logbook. Of note, even when an adequate entrance protocol is implemented, the compliance may remain a challenge, especially when the measures must be frequently repeated or applied by many people (Alarcón et al., 2021).

In our study, semen and small materials or drugs deliveries were reported on nearly all sites (99% and 98%, respectively) over the one-month period, followed by bags (87%) and equipment (61%). Since some equipment from repair or maintenance services might have been previously used in other pig sites, these could represent a higher risk of introducing pathogens in herds unless they are sanitized before entering the unit or kept within a specific reception room allowing sanitization or down time period (Dee et al., 2004; Pitkin et al., 2009b). Unfortunately, on the majority of the sites these precautions were not always taken to reduce the risk associated with the introduction of supplies and equipment. Even new materials can pose a risk of pathogen introduction as they can get contaminated during the delivery process (Pitkin et al., 2009a). Although the null hypothesis was not rejected, feed bags were delivered outside the breeding unit about 1.5 times more often on site having ≥ 2 bags deliveries per month (72%) compared to sites with one delivery (44%) (Table 8), which would warrant further investigations. Producers might have a greater awareness of the risk associated with the introduction of feed bags, particularly in view of its role in the transmission of PED, which was reported following the introduction of PED in 2013 in North America (Scott et al., 2016). The lack of appropriate storage space or the difficulty in sanitizing these bags might also contribute to the decision of delivering outside the unit.

A median and maximum number of pig movements of 5 and 16, respectively, were observed over the one-month period. Pig movements are associated to the spread of PRRSV (Amirpour Haredasht et al., 2017; Makau et al., 2021; Galvis et al., 2022), *Salmonella* (Lurette et al., 2011), and PED virus (Machado et al., 2019). Among pig movements, replacement gilts certainly represent a risk factor for disease introduction into breeding herds (Bottoms et al., 2013) and this risk should not be overlooked considering that 62% of the 84 sites declared at least one gilt introduction over the one-month period. Even when tested negative for pathogens in their herd of origin, gilts can become infected before their departure, or during transport, underlining the importance of an appropriate quarantine in all herds. Unfortunately, most sites reporting gilt introduction in our study did not use a quarantine. Both the location of the quarantine and the type of pig flow could influence the risk of disease introduction into breeding herds as previously reported (Bottoms et al., 2013). These characteristics were not associated with the frequency of gilt introduction in our study. The implementation might rather depend on the presence of infrastructure available on the site or on the global management of gilts since vertically integrated systems can have their own gilt development unit(s). Unsurprisingly, the expedition of weaned piglets and culled sows were the most frequent types of pig movements among sites and on most sites, delivery men were not allowed within the unit to help with pig loading which should decrease the risk of pathogen introduction through contaminated fomites (Otake et al., 2002; Pitkin et al., 2009a). Considering the widespread application of this measure in studied sites, no conclusion can be drawn regarding the association with the frequency of pig movements. The

frequency of pig movements, except for gilts, was not associated with a back-to-back transportation or the use of a loading dock with a truck washed and disinfected; these were used in variable proportions of herds depending on pig categories. This suggests that other factors could have driven their implementation as the type of transport companies involved or the health status of incoming pigs or of the receiving location.

Almost all (98%) sites had service vehicles entering the breeding sites over a one-month period, feed mill and rendering trucks being the most frequent; the large majority of sites experiencing at least 4 entries from at least 3 service providers. Pathogens may persist on truck wheels and be conveyed over several tens of kilometers, and the frequency of vehicle entries increases the risk of contamination of the CAZ (Dee et al., 2002; Dee et al., 2004). In the absence of a rigorous entrance protocol, pathogens may be introduced through contaminated fomites (Otake et al., 2002). It has been recommended that service vehicles use a different path from the one used by staff (Levis and Baker, 2011). Our results showed that only 5% of the sites implemented this measure for feed mill deliveries, which according to a model on transmission of PED would be responsible for 43% of the PED virus introduction in swine breeding herds (Galvis et al., 2022). The use of a wheel bath for trucks at the entrance of the pig farm could reduce contamination of the wheels before entering the premises (Pinto and Urcelay, 2003; Zhang et al., 2013). This measure was not observed in our sites and would be very difficult to implement in winter under our latitude besides a questionable efficacy when disinfectant product becomes inactive or when adhering material prohibits further disinfection.

Manure vacuum and rendering trucks might represent a higher risk for pathogen transmission compared to other service vehicles, since they both carry material susceptible to be contaminated with several pathogens (Levis and Baker, 2011). On sites using shared equipment, this equipment was more likely to be washed before entrance on sites with more than one vacuuming of the manure tank compared to sites with only one. Studies have reported an association between rendering and some infectious diseases (Rose and Madec, 2002; Lambert et al., 2012; Preis et al., 2022). Rendering vehicles can visit a large number of sites on a daily basis, including sites with increased swine mortality, which could increase the risks of contamination (Levis and Baker, 2011; Nathues et al., 2018) and therefore it is recommended that they be denied access to the site. Fortunately, our results showed that rendering was more frequently performed at ≥ 100 m from the pig unit among sites reporting more than one rendering truck visits compared to others. In addition, carcasses should either be transferred in the rendering bin at the end of the day, or all staff should observe a rigorous re-entrance protocol to prevent the introduction of contaminated fomites. Some sites in our study had a freezer within the unit which helps reduce the in and out to the rendering bin.

Most sites investigated were located in Montérégie and Chaudière-Appalaches which have the highest pig site density in Quebec (CDPQ, 2021). This density increases the risk of environmental contamination and the spread of pathogens over areas that can occur through multiple contaminated fomites and aerosols. Aerosol-associated risks can be mitigated with an air filtration system, which was observed in only 5% of the sites. Significant investment may have precluded the installation of air filters into commercial breeding sites. It is surprising that in half of the installations with an air filtration system, the system either was not functional, due to mechanical problems, on standby awaiting maintenance or simply turned off. On 3 sites, manure provided by other pig sites was spread at less than 100 m from a unit. This could be a risk of disease introduction, since many enteric pathogens can survive in manure (Guan and Holley, 2003; Fongaro et al., 2014; McCarthy et al., 2015; Tun et al., 2016; Gallien et al., 2018). Whereas dogs and cats were not authorized within the breeding units, the presence of wild birds was reported on some sites (8%), and this was correlated with an absence of wire mesh bird screen in all air inlets. Since wild birds can carry pathogens transmissible to pigs such as coronaviruses (Mitek and Blicharz-Domańska, 2018) and influenza (Walters, 2014), adequate biosecurity

measures should be present (Alarcón et al., 2021).

The risk of introducing pathogen into breeding herds depends on both the frequency of contacts and the presence of biosecurity measures that reduce the risks associated with each contact. As the number of contacts increases, the importance of biosecurity measures should be emphasized accordingly. However, despite the few associations previously discussed, our results suggest that the implementation of biosecurity measures on swine breeding sites was not influenced by the frequency of contacts. In addition, biosecurity measures were suboptimal in several sites, suggesting that other factors such as lack of knowledge, insufficient time, lack of infrastructure or cost may represent barriers to their adoptions. Alarcon et al. (2014) clearly identified that several of these factors influence the decision-making process for disease control. Considering that only sites with recent PRRS introduction were included in our study, it is also possible that their respective biosecurity measures were not representative of all breeding sites in Quebec. Nevertheless, our results are relevant for identifying areas for biosecurity improvement in our study area given that these sites all experienced a pathogen introduction. Monitoring biosecurity measures would be helpful to determine whether there has been an improvement over time since our data were collected some years ago.

Our study showed, as expected, that the number of staff and visitors and frequency of live pig movements increased with the sow inventory. These factors most probably contribute to the risk of disease introduction, and thus are likely among the underlying reasons driving the reported association between herd size and a wide variety of diseases (Gardner et al., 2002; Hybschmann et al., 2011; Velasova et al., 2012). Our results also showed that breeding sites with a ≥ 4 -week batch farrowing had less live pig movements than sites with < 4 -week batch farrowing, which is related to a decrease in the number of 5 kg piglet and culled sow movements, with weaning performed only once a month. Indeed, some of the objectives of a batch farrowing system are to decrease pig movements to reduce the risk of introduction and to improve all-in-all-out production for a better control of diseases (Bown, 2006).

Different methods were used for data collection. The reliability of data obtained from the interviews (e.g., number of deliveries, entries of service vehicles, etc.) is expected to be lower due to recall bias compared to those obtained from a logbook or traceability system. Thus, the association found between the number of visitors and the presence of a logbook might be partly attributable to a more complete reporting of visitors for sites with logbooks. Nevertheless, the questionnaire only covered the one-month period prior to a recent PRRSv introduction, which decreases the possibility of a memory bias compared to a longer period. We cannot ascertain the absence of recall bias. However, in the context of a PRRSv introduction, the interviewee might have been inclined to recall more precisely some contacts, being in search of possible contamination sources.

Except for live pig transportation, the number of contacts from the same source obtained by interviews was truncated at ≥ 4 for feasibility, limiting the ability to precisely describe the total number of contacts per site. Likewise, only persons and material deliveries into the breeding unit were considered, which could have underestimated the total number of contacts occurring on premises with multiple pig units. Data on biosecurity measures were collected using a categorical scale and the value was determined globally over the one-month period. More precision could have been observed if each biosecurity measure had been evaluated for single contacts. In addition, biosecurity measures were based on auto-reporting and no evaluation or audit were performed at the farm to confirm the declarations, it is possible that their application was overestimated due to social desirability bias.

5. Conclusion

A high degree of contacts was observed in swine breeding sites. Although this level of contact was suspected, very few published data

are available, and most are anecdotal. The frequency of contacts was generally not associated with biosecurity measures, suggesting that a high frequency of contacts is not necessarily perceived as a reason to improve biosecurity. The number of staff, visitors and live pig movements was influenced by sow herd inventory. Our results might be useful for further studies on modelling disease risk in breeding herds located in similar production systems.

Ethics Statement

The protocol was reviewed and approved by the Ethic Committee of the Faculty of Veterinary Medicine, Université de Montréal, Certificate Number CERES-14-128-D.

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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