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Biosecurity Practices and Production Characteristics Associated with High-Pathogenicity Avian Influenza in Québec Duck Farms: A 2022–2023 Case-Control Study

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Abstract

The high-pathogenicity avian influenza (HPAI) epidemic that began in 2021 in Canada continues to affect poultry populations, resulting in the loss of millions of birds through 2026. To investigate farm-level risk factors associated with HPAI infection, a case–control study was conducted on 28 commercial duck farms in Québec, Canada. Farms completed an adapted Biocheck.Ugent questionnaire on biosecurity practices and production characteristics. Associations with HPAI status (case or control) were assessed using Fisher's exact and Wilcoxon rank-sum tests, and multivariate patterns were explored using multiple correspondence analysis (MCA) followed by hierarchical clustering on principal components (HCPC). Case farms had higher annual duck production and were more often part of an integrated production network than control farms. They also reported less frequent wildlife and structural inspections and more often purchased or produced ducks for force-feeding. MCA showed clear separation between case and control farms, driven by differences in biosecurity practices (e.g., hygiene practices, downtime) and farm characteristics (e.g., flock size, indoor housing type). HCPC identified three farm clusters, including one composed exclusively of case farms, characterized by force-feeding production and weaker between-barn hygiene. These findings suggest that production structure and biosecurity profiles should be considered when identifying farms at higher risk of HPAI infection and developing targeted prevention and control strategies.

Keywords: avian influenza; high-pathogenicity avian influenza; biosecurity; case–control; risk factors; duck farms; cluster analysis

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1. Introduction

High-Pathogenicity Avian Influenza (HPAI) represents one of the most important infections affecting bird populations, causing high mortality and morbidity and severe economic losses [1,2]. HPAI also poses a threat to humans, since some strains of HPAI have human pandemic potential [3]. Places where people come into close contact with birds, such as live bird markets and commercial farming, are potentially at higher risk for zoonotic

transmission [4,5]. This is particularly a concern with the H5N1 HPAI currently circulating in many countries, including Canada.

Ducks play a particular epidemiological role in the ecology of avian influenza viruses: they can act as asymptomatic carriers of avian influenza (AI) virus, and can survive HPAI infections, contributing to virus dissemination within and between farms [6,7]. In Canada, recurrent H5N1 outbreaks have affected multiple poultry sectors, including the duck industry. From December 2021 to August 2026, around 17 million domestic birds have been impacted [8]. Some studies suggest that farms with lower biosecurity and greater exposure to wild birds can be more vulnerable to AI [9]. Biosecurity is recognized as a cornerstone of HPAI prevention and control. To prevent economic losses and risks to human health, biosecurity measures are implemented on poultry to reduce the risk of pathogen introduction, establishment, and spread by reducing sources of infection and increasing separation between these sources and healthy flocks [10].

In Québec, duck production includes birds raised for meat and foie gras, with the province being the only producer of foie gras in Canada [11]. The first H5N1 HPAI outbreaks were reported in Québec in April 2022. Between April 2022 and May 2023, 57 commercial poultry premises were affected by avian influenza, including 25 premises involving duck production [12]. These outbreaks brought increased attention to biosecurity in Québec's duck industry. However, empirical data describing farm-level practices and their association with infection risk remain limited. Understanding which measures are associated with farm infection status can help refine prevention strategies and guide the development of targeted interventions. Therefore, the objectives of this study were to (i) describe biosecurity practices implemented on commercial duck farms in Québec during the 2022–2023 HPAI outbreaks, (ii) identify factors associated with the infection status of farms (case or control), and (iii) characterize farm biosecurity profiles based on multivariate analyses.

2. Materials and Methods

2.1. Study Design

This study followed a case–control design including 28 commercial duck farms located in Québec, Canada. All duck farms with more than 300 ducks/geese, considered commercial as per the Canadian Food Inspection Agency's definition [13], and willing to participate in the study were eligible. The objective of this approach was to explore risk factors associated with HPAI infection using a questionnaire referring to practices during the 2022–2023 HPAI outbreaks.

2.2. Selection of Farms

Farms were recruited from lists provided by the Association of Duck and Goose Producers of Québec (Association des éleveurs de canards et d'oies du Québec; AECOQ) and the Ministry of Agriculture, Fisheries and Food of Québec (Ministère de l'Agriculture, des Pêcheries et de l'Alimentation du Québec; MAPAQ). The AECOQ provided a list of its members, while MAPAQ provided a list of registered producers operating farms with more than 300 ducks or geese. Two trained research interns contacted producers to assess their interest in participating in the study.

Of the 50 AECOQ members initially identified, 22 were excluded due to ineligibility ($n = 13$), geographical constraints ($n = 1$), or non-participation ($n = 8$). An additional 19 farms were identified through MAPAQ records, of which 17 were excluded due to production changes or cessation ($n = 5$), geographical constraints ($n = 8$), or non-participation ($n = 4$).

The AECOQ and MAPAQ lists were not considered exhaustive, and additional farms were identified during recruitment. Overall, 36 farms participated in the broader study,

including farms producing meat ducks and breeder ducks. For the present analysis, farms were eligible if information on biosecurity practices during the 2022–2023 HPAI outbreaks was available. Breeder-duck farms were excluded from the present analysis because of their limited number ($n = 4$). Among the eligible meat-duck farms for the present study, 28 were included in the analysis.

2.3. Data Collection

During a single farm visit, participating producers answered a detailed on-farm biosecurity questionnaire referring retrospectively to biosecurity practices implemented in 2022–2023, referring to the HPAI epidemic period. The questionnaire was adapted from the Biocheck.UGent questionnaires (https://biocheckgent.com/en, accessed on 16 September 2026) for duck and goose production, with some modifications to better reflect the Québec production context. In addition, 28 supplementary questions were added to capture biosecurity compliance factors and potential risk factors previously identified in epidemiological investigations of avian influenza outbreaks in Canada [14]. Terminology and response options were revised to ensure clarity and regional relevance and tested with a duck producer. The final questionnaire (in Supplementary Materials) comprised 129 questions divided into sections, covering both external and internal biosecurity aspects: (i) Farm characteristics. (ii) External biosecurity, including A. infrastructure, location, and housing; B. organization of the farm and supply of materials; C. visitors and personnel; D. purchase of ducklings; E. purchase of ducks for force-feeding; F. shipment to slaughter or gavage facilities; G. feed and water supply; and H. manure and carcass removal. (iii) Internal biosecurity, including I. flock management; J. measures between barns; and K. cleaning and disinfection.

Whenever possible, certain answers were validated directly on-site by the interviewer, for instance by checking the presence of other animal species, rodent control measures, barn entrance design or visitor management practices.

2.4. Data Preparation

The research interns completed the online Biocheck questionnaire to obtain the biosecurity scores. The questionnaire responses and corresponding biosecurity scores were entered into SimpleSurvey (https://simplesurvey.com, accessed on 16 November 2026) and exported to an Excel spreadsheet (version 2024; Microsoft Corporation, Redmond, WA, USA), which was subsequently imported into R software (version 4.3; [15]), where all statistical analyses were conducted. Data cleaning procedures were first conducted to verify consistency, coding accuracy, and completeness of responses. Descriptive summaries were used to identify missing values and variable distributions.

Preparation of variables for multivariate analyses (MCA and HCPC) involved additional filtering steps. Variables showing near-constant distributions (≥75% of responses concentrated in a single category; e.g., 89% of respondents reported not having the site fenced) were excluded to avoid undue influence on dimension construction [16]. For the remaining variables included in the MCA, 11 missing values (1% of all responses) were imputed using multiple correspondence analysis-based imputation implemented in the missMDA package (version 1.20) [17].

To ensure balanced category representation and interpretability in the MCA, selected variables were recategorized prior to analysis. A total of 18 categorical variables were collapsed by merging conceptually similar response categories to reduce sparsity and avoid rare modalities disproportionately influencing the construction of the MCA dimensions. In particular, ordinal response options (e.g., never, sometimes, always) were collapsed into binary categories (e.g., yes and no) and closely related structural or management categories were grouped based on biological relevance and biosecurity principles. For

example, different forms of visitor notification (logbooks located at barn level or at the farm entrance) were collapsed into a single category reflecting the presence of a visitor logbook, as both categories represent almost the same concept.

2.5. Statistical Analyses

Biosecurity scores generated by the Biocheck.Ugent assessment tool were compared between case and control farms using the Wilcoxon rank-sum test. Overall biosecurity scores, external and internal biosecurity subtotals, and individual section scores were evaluated. Scores ranged from 0 to 100, with higher values indicating higher biosecurity levels. Sections E (Purchase of ducks for force-feeding) and J (Measures between barns) were not statistically compared because of the limited number of applicable farms. Section D (Purchase of ducklings) was analyzed using available observations only. Results were summarized as medians and interquartile ranges.

Data were analyzed to identify statistical association between farm infection status (case vs. control) and farm characteristics and biosecurity practices. Univariate analyses were first performed to screen variables potentially associated with farm infection status. For univariate analyses, original response categories were retained. Categorical variables were summarized as frequencies and percentages and tested using Fisher's exact test. Continuous variables were assessed for normality with the Shapiro–Wilk test; as none followed a normal distribution, comparisons were conducted using the Wilcoxon rank-sum test (Mann–Whitney U), a nonparametric test for non-normal data or small samples. To facilitate interpretation, selected quantitative variables (e.g., number of production cycles, flock size, building age) were also categorized into quartiles and re-evaluated as categorical variables.

An exploratory multivariable analysis was initially attempted using a logistic regression method designed for small sample sizes, rare events, and complete or quasi-complete separation (Firth's penalized logistic regression). Variables associated with farm HPAI status at $p < 0.20$ in univariate analyses were considered eligible for multivariable modeling. Prior to model building, correlations among candidate variables were assessed using Cramér's V to reduce collinearity, and a subset of non-redundant variables was selected for inclusion.

2.5.1. Multiple Correspondence Analysis

The MCA is a method used to analyze survey data and visualize similarities among individuals and associations among categorical variables [18]. The MCA projects categories and individual observations (e.g., farms, people) into a low-dimensional space where proximities between points reflect associations [19].

An MCA was conducted with the FactoMineR (version 2.11) and factoextra (version 1.0.7) packages to explore associations between farms and biosecurity practices. The farm infection status (Case vs. Control) was included as a supplementary qualitative variable, meaning it did not influence the construction of the axes but was projected afterward to illustrate interpretation [18]. Five dimensions were retained for interpretation based on eigenvalue decomposition (which quantifies how much variance each dimension explains) and the visual inspection of a scree plot (which displays the diminishing returns of successive dimensions). The contributions of variables and categories to the first two principal dimensions were examined, and top-contributing variables were visualized using biplots with confidence ellipses around case and control groups. Categories with higher contributions played a greater role in defining the corresponding dimension, meaning that they contributed more strongly to the contrasts captured by that axis and to the positioning of farms along it. Confidence ellipses can be drawn around the categories of the supplemen-

tary categorical variable and allow visualization of whether or not two categories differ significantly [18].

2.5.2. Hierarchical Clustering on Principal Components

To identify farm typologies based on biosecurity practices and farm characteristics, a HCPC was performed on the results of the MCA using the FactoMineR package [20]. Hierarchical clustering was conducted using Ward's method with Euclidean distance. Initially, each farm was treated as an individual cluster. At each step of the algorithm, the two clusters whose merger resulted in the smallest increase in total within-cluster inertia were combined. This iterative method continued until the growth of within-cluster inertia was minimal [18].

The optimal number of clusters was determined automatically by the HCPC algorithm based on the gain in within-cluster inertia, which reflects the balance between cluster compactness and separation. The categories describing each cluster correspond to the categories of the original categorical variables. Their association with cluster membership is assessed using a statistical test based on the hypergeometric distribution. Under the null hypothesis, the proportion of a given category within a cluster is expected to be equal to its proportion in the overall dataset [18]. Categories were considered overrepresented in a cluster when their observed frequency within that cluster was significantly higher than expected under this null hypothesis. Categories that are significantly associated with a cluster are identified according to ascending p values. For each category, a v-test (test value) was computed, corresponding to a quantile of the standard normal distribution associated with the p value. Positive v-test values indicate overrepresentation of a category in a given cluster, whereas negative values indicate underrepresentation. A v-test value greater than 1.96 indicates statistical significance at the 0.05 level [18].

Finally, associations between cluster membership and farm infection status (case or control) were evaluated using Fisher's exact test, with statistical significance assessed at a two-sided α level of 0.05.

2.6. Ethics Statement

A confidentiality agreement was signed to protect the data collected. Only data that do not allow individual farms to be identified are presented in the following sections.

3. Results

Questionnaires were completed by farmers from 14 control farms and 14 case farms. Farms were visited between 20 June 2024 and 26 March 2025 and were located in five regions of Québec: Centre-du-Québec, Estrie, Montérégie, Capitale-Nationale, and Chaudière-Appalaches.

3.1. Biocheck Biosecurity Scores

The comparison of Biocheck.Ugent biosecurity scores between case and control farms revealed no statistically significant differences in overall biosecurity scores, external and internal biosecurity subtotals, or individual section scores (Figure 1). The median overall biosecurity score was 66.5 (IQR: 51.8–70.0) for case farms and 65.0 (IQR: 60.0–69.0) for control farms ($p = 0.68$). Although median overall scores were similar, case farms showed greater variability in biosecurity scores. For instance, the largest numerical difference in median scores was observed for the depopulation of ducks and geese section, with a median of 79.5 (IQR: 42.3–90.0) for case farms compared with 90.0 (IQR: 71.0–90.0) for control farms ($p = 0.16$). However, none of the individual section comparisons reached statistical significance. Sections E (Purchase of duck for force-feeding) and J (Measures

between barns) were excluded because of the limited number of applicable farms. Section D (Purchase of ducklings) included eight case farms and 14 control farms.

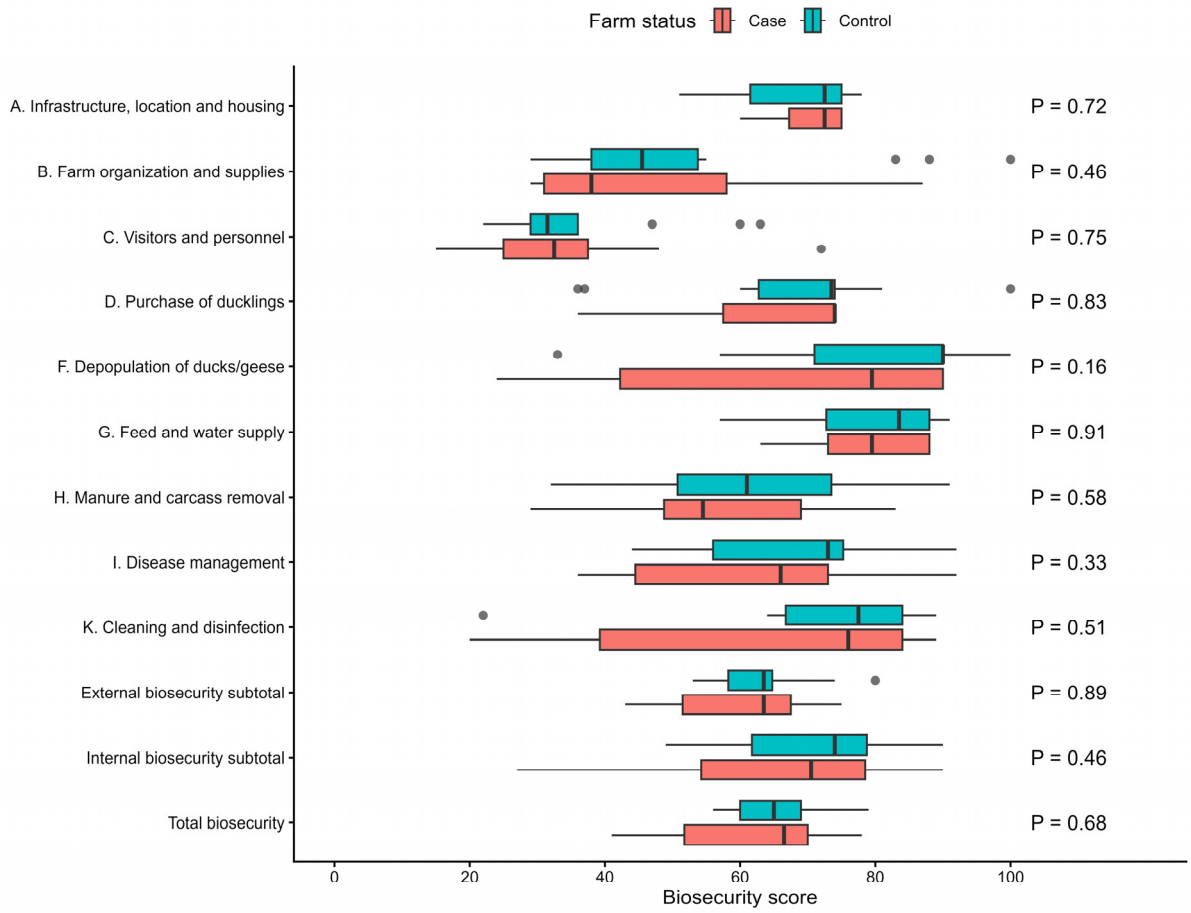

Figure 1. Comparison of Biocheck.Ugent biosecurity scores between high-pathogenicity avian influenza (HPAI) case farms ($n = 14$) and control farms ($n = 14$) in Québec, Canada. Boxplots represent the median and interquartile range (IQR), with whiskers extending to values within 1.5 times the IQR. Outliers are represented by the dots. Sections E (Purchase of force-fed ducks) and J (Measures between barns) were excluded because of the limited number of applicable farms. Section D (Purchase of ducklings) included eight case farms and 14 control farms.

3.2. Univariate Analysis

Results from the univariate analysis are presented following the sections of the questionnaire used. Variables with a $p < 0.05$ are presented in Table 1, while the complete univariate results are provided in Supplementary Materials Tables S1–S13. Variables with $p < 0.20$ were considered eligible for multivariable modeling, as described below.

Table 1. Selected farm-level characteristics and biosecurity practices significantly associated with highly pathogenic avian influenza (HPAI) infection status. The table presents the distribution of case and control farms for variables with a univariate p value < 0.05 based on Fisher's exact test, as well as comparisons of medians for numerical variables using the Wilcoxon rank-sum test.

Categorical Variables	No. of Case Farms (%)	No. of Control Farms (%)	Univariate p Value
Are you part of an integrated network?			0.01
No	1 (7.1)	8 (57.1)	
Partially	5 (35.7)	1 (7.1)	
Yes	8 (57.1)	5 (35.7)	

Table 1. *Cont.*

Categorical Variables	No. of Case Farms (%)	No. of Control Farms (%)	Univariate *p* Value
At what frequency do you inspect your barns to assess integrity for wildlife and avoid water infiltration?			0.02
Once a year	5 (35.7)	0 (0.0)	
More than once a year	8 (57.1)	14 (100.0)	
When I have a problem	1 (7.1)	0 (0.0)	
Do you rear ducklings/goslings at the same location?			0.02
No	6 (42.9)	0 (0.0)	
Yes	8 (57.1)	14 (100.0)	
Do you purchase/produce pre-reared ducks for force-feeding?			0.03
No	7 (50.0)	13 (92.9)	
Yes	7 (50.0)	1 (7.1)	
Numerical variables	Median Case farms	Median Control farms	*p* value (Wilcoxon rank-sum)
Number of ducks/geese On an annual basis	138,000	9700	0.01
Number of production cycles (flocks)/year	13.0	4	0.01

3.2.1. Section Introduction—Farm Characteristics

Case farms had a significantly higher median number of production cycles per year (13 cycles vs. 4 cycles; $p = 0.014$) and a significantly higher median annual duck count (138,000 vs. 9700 ducks, $p = 0.01$) compared with control farms (Supplementary Table S1). No differences were observed between case and control farms for the number of full-time employees or building age. Farms that were part of an integrated production network were significantly more likely to be classified as HPAI case farms (57% vs. 36%; $p = 0.01$; Table 1). No significant associations were observed between HPAI status and family access to birds, slatted flooring, or contact with other poultry farms (Supplementary Table S2).

(i) External biosecurity

3.2.2. Section A—Infrastructure, Location and Housing

A statistically significant difference was observed for wildlife and structural inspection frequency, with control farms reporting more inspections than case farms (100% vs. 57%; $p = 0.02$; Table 1). A borderline association was observed for rodent control practices ($p = 0.05$; Supplementary Table S3). The same percentage of case and control farms used professional pest control services (11/14, 78.6% vs. 11/14, 78.6%). Among the six farms that did not use professional pest control services, all three case farms reported having no rodent control program (3/14; 21.4%), whereas all three control farms implemented their own rodent control measures (3/14; 21.4%).

3.2.3. Section B—Organization of the Farm and Supply of Materials

The presence of a written biosecurity plan, biosecurity training of owners or employees, the reported restrictions on non-essential vehicle entry, separation between clean and dirty areas, disinfection of vehicle wheels, and sharing of equipment within or between farms did not differ between case and control farms.

3.2.4. Section C—Visitors and Personnel

No statistically significant associations were found between visitor and personnel-related biosecurity measures and HPAI infection status. Overall, visitor notification procedures, access restrictions, hygiene measures, and personnel practices, as reported by the farmers, were similar between case and control farms (Supplementary Table S5).

3.2.5. Section D—Purchase of Ducklings

A statistically significant association was observed for rearing ducklings on the same farm location ($p = 0.02$). All control farms reported rearing ducklings on the same site, whereas 43% ($n = 6$) of case farms reported not raising ducklings (Table 1). No significant differences were observed between case and control farms for other duckling-related practices, including the use of the same supplier over the previous two years, delivery order of ducklings, requests for proof when purchasing from other farms, use of disposable transport boxes, or verification of cleanliness and disinfection of containers and transport vehicles (Supplementary Table S6).

3.2.6. Section E—Purchase of Ducks for Force-Feeding

A statistically significant association was found between purchasing or producing ducks for force-feeding and HPAI infection status (Table 1). Case farms were more likely to report purchasing or producing ducks specifically for force-feeding compared with control farms (50% vs. 7%; $p = 0.03$). No statistically significant differences were observed between case and control farms for other practices related to duck sourcing, delivery order, verification of origin, transport and container cleaning and disinfection, employee sharing, or crate management (Supplementary Table S7).

3.2.7. Section F—Shipment to Slaughter or Gavage

No statistically significant associations were observed between catching-related biosecurity practices and HPAI infection status. Practices related to transport vehicle and crate sanitation, cleaning and disinfection of loading areas and barn entrances, and catching practices were similar between case and control farms (Supplementary Table S8).

3.2.8. Section G—Feed and Water Supply

No statistically significant associations were observed between feed- and water-related biosecurity practices and HPAI infection status. Practices related to feed delivery, supplier access to barns, feed truck sanitation, feed spillage management, water treatment, and drinking water monitoring were similar between case and control farms (Supplementary Table S9).

3.2.9. Section H—Manure and Carcass Removal

No statistically significant associations were observed between manure or carcass management practices and HPAI infection status. Most practices related to manure storage and removal, carcass storage conditions, disposal methods, and hygiene measures during dead bird handling were similar between case and control farms (Supplementary Table S10).

(ii) Internal biosecurity

3.2.10. Section I—Flock Management

No statistically significant associations were observed between flock management practices and HPAI infection status in univariate analyses. Practices related to disease monitoring, frequency of flock checks, removal of dead birds, stocking density, seasonal ad-

justment of animal density, multi-age flock management, and employee movement between flocks were similar between case and control farms (Supplementary Table S11).

3.2.11. Section J—Measures Between Barns

No statistically significant associations were observed concerning between-barn biosecurity practices and HPAI infection status in univariate analyses. Other measures related to barn access, the reported use and cleaning of corridors and anterooms, footbath use, hand hygiene, and barn-specific clothing, footwear, and equipment were similar between case and control farms (Supplementary Table S12).

3.2.12. Section K—Cleaning and Disinfection

No statistically significant associations were observed between cleaning and disinfection practices and HPAI infection status in univariate analyses. Practices related to barn cleaning and disinfection, use of detergents, drying procedures, verification of cleaning efficacy, cleaning of feeding and watering systems, and material sanitation were similar between case and control farms (Supplementary Table S13).

3.3. Multivariable Analysis

Six variables with $p < 0.05$ and ten additional variables with $0.05 \leq p < 0.20$ in the univariate analyses were considered eligible for exploratory multivariable analysis. Variables with $p < 0.05$ were described in the sections above, whereas variables with $0.05 \leq p < 0.20$ included indoor housing on litter, indoor housing in collective cages, rodent control program, regular observation of migratory birds, fresh litter provision for each batch, storage of clean bedding on the farm before use, use of a contractual catching crew, manure and litter storage location, presence of multiple duck barns, and duration of downtime between production cycles (Supplementary Tables S1–S13). However, the fitted Firth-penalized logistic regression models produced unstable estimates and large standard errors. Given these limitations, the multivariate results were not considered reliable for interpretation and are not presented.

3.4. Results of Multiple Correspondence Analysis

After applying the variable filtering procedures described in the material and methods section, the final MCA dataset included 28 farms and 40 categorical variables (39 active and one supplementary variable).

The MCA yielded a 27-dimensional solution, with Dimensions 1 and 2 together explaining 35% of the total variance (Dimension 1 = 22.2% and Dimension 2 = 12.7%). The variables contributing most strongly to Dimension 1 were those related to the presence of a poultry health management program, verification of cleaning efficacy, housing system (slatted floors and litter use), and wildlife presence indicators (e.g., corvids, pigeons; Figure 2). For Dimension 2, the highest contributions were associated with practices related to buying ducks for force-feeding, housing configuration (e.g., single vs. multiple barns), cleaning and disinfection procedures, equipment introduction measures, visitor biosecurity requirements, downtime between production cycles, and being part of an integrated production network (Figure 2).

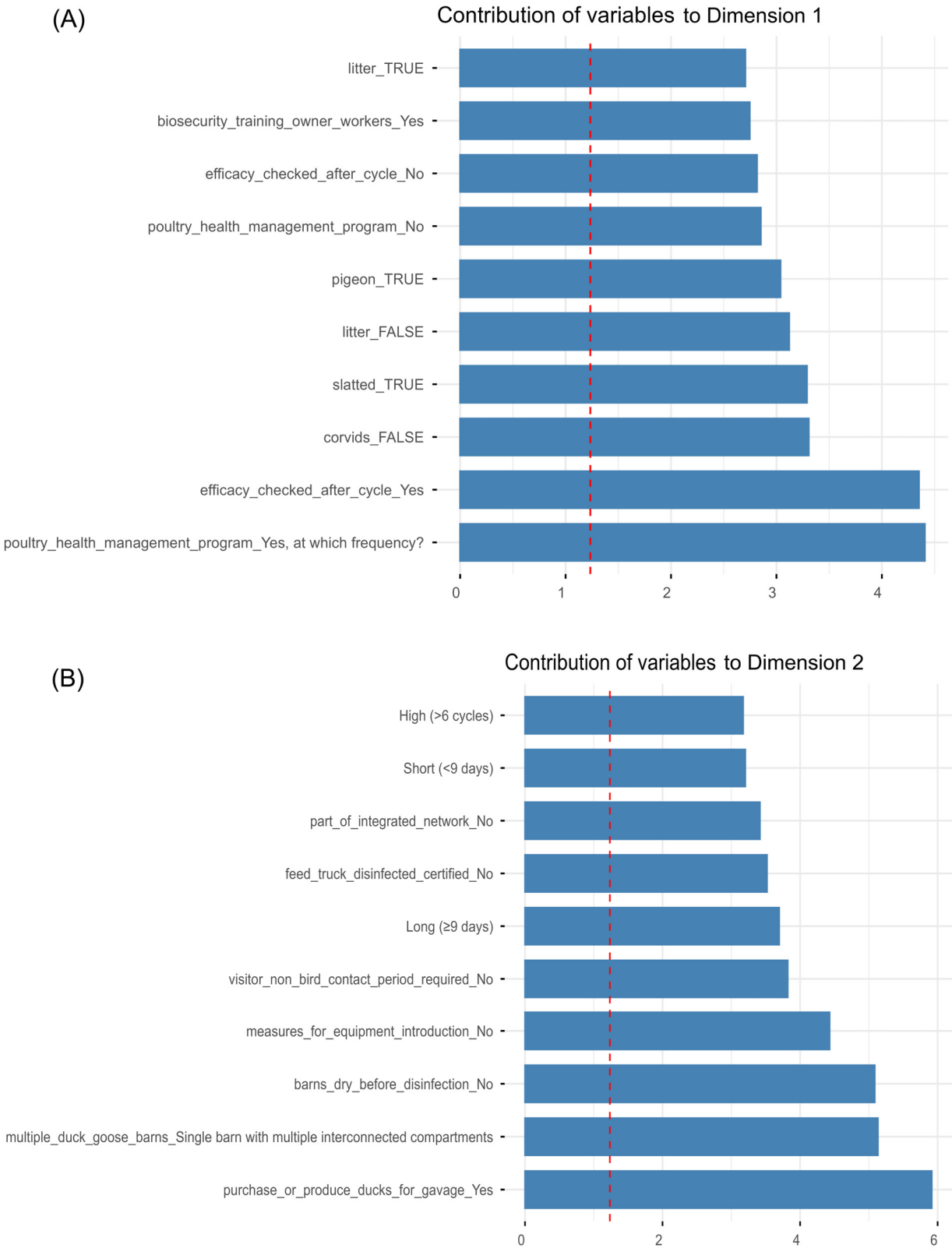

Figure 2. Contribution of categorical variables to the first two dimensions of the multiple correspondence analysis (MCA). (**A**) Dimension 1. (**B**) Dimension 2. Bars represent the percentage contribution of each category to each dimension. The red dashed line indicates the expected average contribution. Categories above this threshold were among the strongest contributors to the construction of the dimension.

Ellipses plotted around the barycenters of the supplementary variable (farm status: Case vs. Control) indicated clear separation between case and control farms along the first two dimensions (Figure 3). Case farms were primarily positioned toward the positive side of Dimension 1 and showed higher association with less strict biosecurity indicators (e.g., no measures for equipment introduction, absence of visitor non-contact period, no feed truck disinfection), whereas control farms clustered toward the negative side of Dimension 1, aligning with stronger biosecurity practices. Overall, this position suggests that biosecurity organization, production structure, and hygiene processes differed between HPAI-infected and non-infected farms (Figure 3).

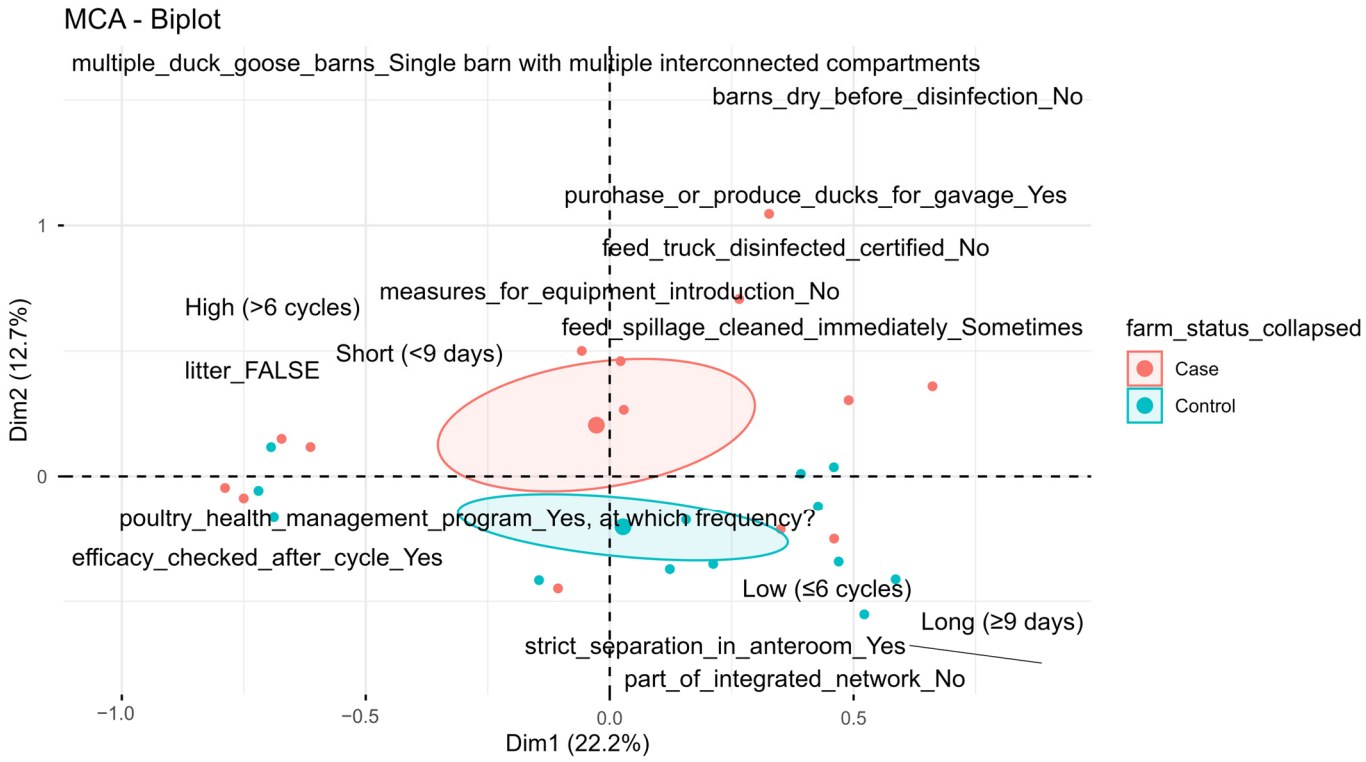

Figure 3. Multiple correspondence analysis biplot of biosecurity practices and production characteristics in 28 commercial duck farms. The first two dimensions reveal separation between HPAI case and control farms. Case farms cluster toward categories associated with higher production intensity and less strict biosecurity measures, whereas control farms align with longer downtime periods, stricter anteroom separation, and non-integrated production systems. Ellipses indicate 95% confidence regions around group barycenters.

3.5. Results of Hierarchical Clustering on Principal Component

Three distinct clusters were identified based on their MCA coordinates. These clusters were well separated on the map, indicating structural and biosecurity-related differences among groups (Figure 4). Cluster 1 included eight farms with an equal number of cases and controls (4 and 4, respectively). This cluster was characterized by large farms (≥4200 ducks), barns with slatted floors, a poultry health management program in place, and a downtime period of less than nine days between production cycles (Table 2).

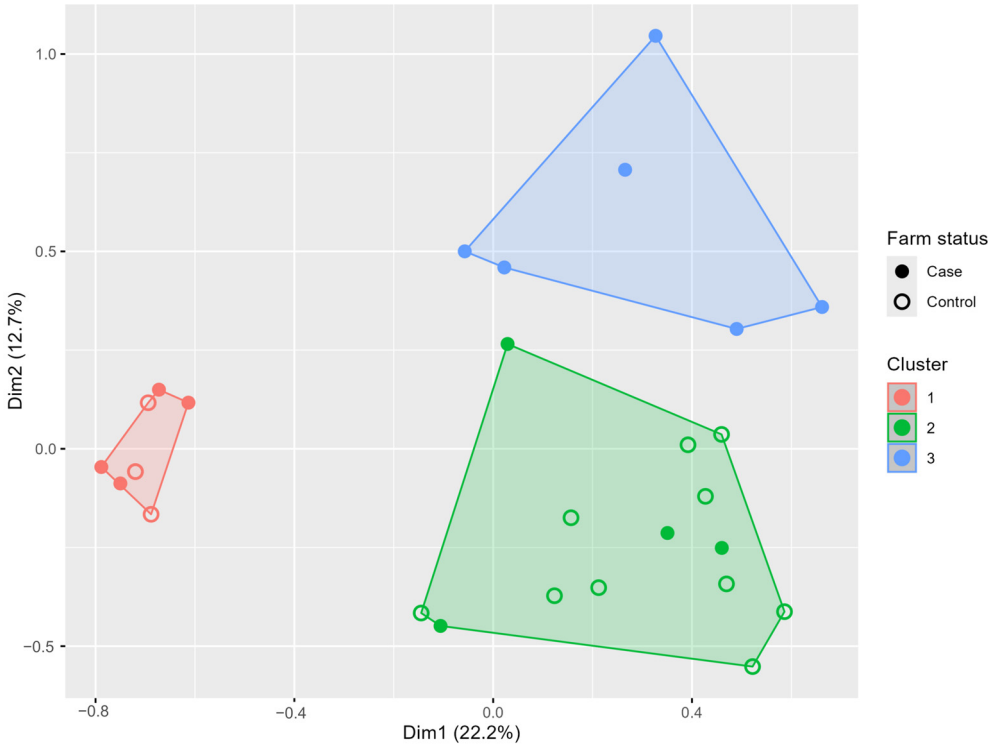

Figure 4. Cluster plot derived from hierarchical clustering on principal components based on the MCA. Farms are projected onto the first two MCA dimensions and grouped into three clusters. Points represent individual farms, with open circles indicating control farms and filled circles indicating case farms. The spatial separation of clusters reflects distinct biosecurity profiles and production structures among farms. Cluster 1 has 8 farms (4 control and 4 cases), Cluster 2 has 14 farms (10 control and 4 cases), and Cluster 3 has 6 case farms.

Table 2. Categories that were associated with each cluster. Modalities shown are those significantly overrepresented within each cluster (v-test > 0; $p < 0.05$). Percentages correspond to the proportion of farms within each cluster presenting the given category (Mod/Cla). Only positively associated categories are displayed.

Cluster (n)	Variable (Category)	% Within Cluster (Mod/Cla)	v-Test
Cluster 1 ($n = 8$)	Efficacy of cleaning/disinfection checked after cycle = Yes	100%	4.04
	Poultry health management program = Yes	100%	4.04
	Slatted floors = Yes	100%	4.04
	Litter = No	100%	3.53
	Larger farms (>4200 ducks)	100%	3.30
	Short downtime after cycle (<9 days)	100%	3.08
	Feed spillage cleaned immediately = Always	100%	2.66
	Part of an integrated network	100%	2.25
Cluster 2 ($n = 14$)	Long downtime after cycle (≥9 days)	85.7%	4.14
	Smaller farms (≤4200 ducks)	85.7%	3.70
	Litter = Yes	85.7%	3.30
	Low number of production cycles (≤6)	85.7%	3.30
	Not part of an integrated network	64.3%	3.62
	Strict separation in anteroom = Yes	57.1%	3.30
	Biosecurity training (owner/workers) = Yes	71.4%	2.53
	Control farms	71.4%	2.14

Table 2. *Cont.*

Cluster (n)	Variable (Category)	% Within Cluster (Mod/Cla)	*v*-Test
Cluster 3 ($n = 6$)	Case farms	100%	2.65
	Purchase/produce ducks for force-feeding = Yes	83.3%	2.96
	Hand washing/disinfection in barn = No	83.3%	2.71
	Feed spillage cleaned immediately = Sometimes	83.3%	2.71
	Certified feed truck disinfection = No	66.7%	2.58
	Visitor non-bird contact period required = No	83.3%	2.48
	Measures for equipment introduction (UV-disinfection, alcohol disinfection, soap and water) = No	83.3%	2.48
	Efficacy check after cycle = No	100%	2.13

Cluster 2 included 14 farms and was composed predominantly of control farms (10 control farms and 4 case farms). Farms in this cluster tended to be smaller, were not part of an integrated production network, had strict anteroom separation between dirty and clean areas, and had longer downtime periods (≥9 days) after production cycles (Table 2).

Cluster 3 consisted exclusively of case farms (6 cases, 0 controls). This cluster was characterized by farms that purchased or produced ducks for force-feeding, and where visitors/personnel did not perform hand washing or sanitation upon entering each barn (Table 2).

Farm-status distribution (case vs. control) differed significantly across the three clusters identified by the HCPC (Fisher's exact test, $p = 0.01$), with Cluster 3 composed entirely of case farms (100%, $n = 6$) and Cluster 2 predominantly composed of control farms (71%, $n = 10$).

4. Discussion

This study explored farm-level biosecurity practices and farm characteristics associated with HPAI infection status among commercial duck farms (meat-type) in Québec using a combination of univariate analyses and multivariate exploratory methods. In univariate analyses, case farms were associated with larger flock sizes, the production or purchase of ducks for force-feeding, participation in an integrated production network, and not rearing ducklings or goslings on site. Additionally, case farms reported less frequent inspection of barn integrity for wildlife access and water infiltration. In the clustering analysis, three farm profiles based on farm characteristics and biosecurity measures were identified; two of these profiles were associated with farm infection status. In contrast, comparisons of overall and section-specific Biocheck.Ugent biosecurity scores revealed no statistically significant differences between case and control farms.

In this study, case farms had a significantly higher annual duck count than control farms in univariate analyses, suggesting that larger flock size was associated with HPAI infection status. This finding is consistent with the literature, where larger flocks have previously been identified as being at greater risk of HPAI infection [21].

Farms integrated within production networks were more frequently classified as case farms in the univariate analysis. In addition, farms that were not part of an integrated network were overrepresented in Cluster 2, which was predominantly composed of control farms. These results suggest that production network integration may be associated with increased HPAI risk. One possible explanation is that farms integrated within production networks may experience more movements of animals, personnel, vehicles, and equipment between premises, thereby increasing opportunities for pathogen introduction and spread

when biosecurity is inadequate. Supporting this interpretation, a study conducted on poultry farms in Ontario found that 53% of visitors had contacted other poultry farms within the previous 72 h, and that seven of eight farms had at least one visitor who reported not implementing biosecurity measures. These movements between farms may facilitate the spread of avian influenza viruses [22,23].

Farms involved in force-feeding production systems were more frequently classified as case farms, suggesting that this production context may be associated with increased exposure to AI viruses. Similar findings have been reported in France, where force-feeding (foie gras) production systems were identified as a risk factor for HPAI occurrence [24]. In these systems, ducks are typically reared in large flocks for up to 12 weeks before being transferred to specialized force-feeding units. The movement of ducks from rearing farms to multiple force-feeding units may increase the number of epidemiological links between farms through the movement of personnel, vehicles, crates and live ducks, facilitating virus introduction and spread [25–27]. This interpretation was further supported by the multivariate analysis: Cluster 3, which was composed exclusively of case farms, was characterized not only by the purchase or production of ducks for force-feeding, but also by weaker visitor and personnel hygiene practices. These findings suggest that movement between farms and suboptimal implementation of visitor and personnel biosecurity measures may have contributed to the observed HPAI infection status of these farms.

Hand washing or hand disinfection for visitors and personnel in each barn was not associated with farm status in the univariate analysis. However, lack of hand washing/disinfection in each barn was overrepresented in Cluster 3 (v-test 2.71), which had a higher proportion of case farms. Incomplete hygiene measures among farm visitors, including inadequate sanitation of boots, hands, and clothing, have previously been reported as a risk factor for the introduction of AI virus in commercial poultry farms [28].

In the present study, rearing ducklings on site was associated with control farms. This finding is different from a previous study that reported that the presence of birds of different ages on the same site is a risk factor for avian influenza [5]. However, on-site rearing in our study may have acted as an indicator of fewer animal movements, whereas the risk identified in the literature likely reflects within-farm transmission dynamics related to multi-age production. This association may also have been confounded by production type, as six of the eight farms involved in force-feeding (gavage) did not rear ducklings on site.

The comparison of Biocheck.Ugent biosecurity scores revealed no statistically significant differences between case and control farms, either in overall biosecurity scores or in individual sections. Although these scores provide an overview of farm biosecurity, combined measures may hide weaknesses in specific practices potentially relevant to HPAI introduction. For example, the univariate analysis identified differences in practices related to wildlife access and farm biosecurity management that were not apparent from the comparison of biosecurity scores. Additionally, the MCA and HCPC identified distinct farm profiles based on a combination of biosecurity practices and production characteristics, providing additional information beyond the scores. However, the absence of statistically significant differences in BioCheck.Ugent scores does not necessarily indicate that the scoring system is unable to distinguish biosecurity levels between farms, particularly given the small sample size.

Interpretation of these findings should consider the study limitations. Biosecurity practices were collected retrospectively during a single farm visit, requiring producers to report practices implemented in 2022–2023 during visits conducted in 2024. This approach may have introduced recall bias, as producers may not have accurately remembered the specific biosecurity measures in place during the outbreak period. In addition, because biosecurity practices were self-reported, responses may have been influenced by social

desirability bias, with producers potentially overstating compliance with recommended measures. This bias may have been particularly relevant among case farms, where respondents could have been more reluctant to disclose biosecurity shortcomings following an HPAI outbreak. Consequently, differential reporting between case and control farms may have influenced the observed associations.

Importantly, the reliance on producer-reported information limited our ability to verify whether the reported biosecurity measures were consistently implemented during the study period. Although several responses were verified on-site by interviewers through direct observation of facilities and infrastructure, these observations could not confirm the consistent implementation of practices during the outbreak period. Additionally, the small number of participating farms limited the use of multivariable logistic regression models and reduced statistical power to detect more subtle associations. However, the duck production sector in Québec is limited in size, and the study included a substantial proportion of eligible farms, supporting its relevance within this production situation. In this context, the use of MCA was particularly appropriate, as it allowed exploration of multivariate patterns and farm-level biosecurity profiles without requiring large sample sizes or strict regression assumptions.

Finally, the Biocheck.UGent questionnaire is designed to be species- and production-specific. While this enhances its suitability for duck production systems, it limits the assessment of biosecurity practices in multi-species or multi-production sites, where interspecies interactions may influence disease transmission dynamics. Future studies should also assess whether biosecurity practices changed following the avian influenza outbreaks, and whether these changes were sustained over time. Such research could help identify which practices were most readily adopted after the outbreak and which biosecurity gaps remained.

An important contribution of this study lies in the identification of distinct farm-level biosecurity and production profiles associated with HPAI infection status. The identification of higher-risk farm typologies has practical implications for HPAI prevention. Surveillance and control strategies may benefit from prioritizing farms exhibiting combinations of high connectivity, force-feeding production, and hygiene biosecurity gaps.

5. Conclusions

This study identified differences in farm characteristics and biosecurity practices between HPAI case and control meat-type duck farms in Québec. Case farms were characterized by larger flock sizes and were more frequently associated with integrated production networks and force-feeding production, while on-site rearing of ducklings was more common among control farms. Exploratory multivariate analyses further identified distinct combinations of production characteristics and biosecurity practices associated with farm HPAI infection status, including patterns related to farm connectivity and visitor and personnel hygiene. The results highlight the importance of considering production structure, animal and farm connectivity, and the implementation of biosecurity measures when assessing HPAI exposure in commercial duck farms.

Supplementary Materials: The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/v18111280/s1: Supplementary Table S1: Introduction: Farm characteristics—numerical variables; Table S2: Introduction: Farm characteristics—categorical variables; Table S3: Section A—Infrastructure, location and housing; Table S4: Section B—Organization of the farm and supply of materials; Table S5: Section C—Visitors and personnel; Table S6: Section D—Purchase of ducklings; Table S7: Section E—Purchase of ducks for force-feeding; Table S8: Section F—Shipment to slaughter or gavage facilities; Table S9: Section G—Feed and water supply; Table S10: Section H—Manure and carcass removal; Table S11: Section I—Flock management; Table S12: Section J—Measures between barns; Table S13: Section K—Cleaning and disinfection. Additional

Supplementary Materials include the adapted Biocheck.Ugent questionnaire in French and English, de-identified data, and R scripts used for the univariate analyses, multiple correspondence analysis (MCA), and hierarchical clustering on principal components (HCPC).

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Abbreviations

The following abbreviations are used in this manuscript:

AECOQ	Québec Duck and Goose Breeders Association (Association des éleveurs de canards et d'oies du Québec)
AI	Avian Influenza
HPAI	High-Pathogenicity Avian Influenza
HCPC	Hierarchical Clustering on Principal Components
MAPAQ	Ministère de l'Agriculture, des Pêcheries et de l'Alimentation du Québec
MCA	Multiple Correspondence Analysis

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