Stochastic simulation of the spread of highly pathogenic avian influenza in Cuba

Simulación estocástica de la difusión de influenza aviar altamente patógena en Cuba

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ABSTRACT: Highly pathogenic avian influenza (HPAI) is a major global animal health problem with public health implications. The aim of this study was to estimate the magnitude of the consequences of HPAI virus incursion in Cuba and the effectiveness of the main control measures. A spatial stochastic model was used to represent biological, environmental and human interaction processes involved in transmission. NAADSM (v.3.3.2) tool was used for parameterization and simulation of the outbreak, and R (v. 3.5) tool for statistical analysis of outbreaks. Four hundred sixty-seven poultry farms in 216 scenarios were processed by evaluating 20 parameters associated with probability functions or linear models to represent the epidemic process of Susceptible-Infectious-Removed (SIR) model in the population studied. Simulation showed depopulation was able to stop the outbreak in all scenarios; however, the greatest animal losses were evident in scenarios with poor biosecurity, slow detection speed and reduced movement restriction. Adjacent farms within a 5 km radius had a significantly higher risk of spreading the virus to a greater extent. The model suggests that it is possible to contain the spread of HPAI virus if detection is reached within three days post-infection, and depopulation is completed within six days.

Key words: NAADSM, virus, risk assessment, poultry, modeling, disease.

RESUMEN: La influenza aviar altamente patógena (IAAP) es un importante problema de salud animal a nivel mundial con implicaciones para la salud pública. El objetivo de este estudio fue estimar la magnitud de las consecuencias de la incursión del virus IAAP en Cuba y la efectividad de las principales medidas de control. Se utilizó un modelo estocástico geoespacial para representar los procesos de interacción biológica, ambiental y humana involucrados en la diseminación. Para la parametrización y simulación del brote se utilizó la herramienta NAADSM (v.3.3.2) y la herramienta R (v. 3.5) para el análisis estadístico de los brotes. Se procesaron 467 granjas avícolas en 216 escenarios, mediante la evaluación de 20 parámetros asociados a funciones de probabilidad o modelos lineales para representar el proceso epidémico de Susceptibles Infecciosos Removidos (SIR) en la población estudiada. La simulación mostró que la despoblación logró el cierre del brote en todos los escenarios. Sin embargo, las mayores pérdidas de animales se evidenciaron en escenarios con pobre bioseguridad, lenta velocidad de detección y restricción de movimiento reducida. Los factores más influyentes en la propagación del virus fueron la bioseguridad y la falta de restricción de movimiento efectiva. Las granjas adyacentes en un radio de 5 km tuvieron un riesgo significativamente mayor de propagar el virus en mayor medida. El modelo sugiere que es posible contener la propagación del virus de IAAP si la detección se logra dentro de los tres días posinfección y la despoblación se logra en menos de seis días.

Palabras clave: NAADSM, virus, análisis de riesgo, aves comerciales, modelación, enfermedad.

INTRODUCTION

Infection with high pathogenicity avian influenza viruses (HPAIVs) is a devastating disease of poultry listed by the World Organisation for Animal Health (WOAH) (1). Low pathogenic avian influenza virus (LPAIV) infections, likewise, are no longer reportable to WOAH since 2021 (2), although some strains of the H5 and H7 subtypes have the ability to mutate to HPAIV.

Waterfowl are often repeatedly exposed to and infected with avian influenza viruses with little or no signs of clinical disease. Since 2002, some HPAIVs have become established in certain wild bird popula-

tions, often associated with mortality (3,4). This paradigm shift has been associated with large epidemics, some of them of panzootic magnitude such as those of the H5N1 and H5N8 subtypes from Asian origin (5).

Recently, a significant increase of HPAI outbreaks worldwide has affected domestic and wild birds, and some terrestrial and aquatic mammals. This reflects a change in the epidemiology and ecology of the virus, which poses a threat to animal health, public health, food security and biodiversity (6,7). This situation implies a renewed need to better understand transmission, spread and potential impact of HPAI to improve control and mitigate negative outcomes.

There is growing evidence that HPAIVs are endemic in the avian reservoir (8), adding complexity to their eradication, which allows an increased occurrence of infections. This may imply a higher risk of disease occurrence in countries connected with migratory flyways. Cuba is not only an important wintering area for migratory birds coming from North America, but also a very important staging area for birds moving to other Caribbean islands or birds migrating further south (9). This, together with the existence of important poultry populations, implies conditions for the occurrence of infections with HPAIVs (10), as recently evidenced by the incursion of HPAI H5N1 in zoo birds (11).

Cuba has implemented an avian influenza surveillance program that includes both passive and active components according to the requirements of the Terrestrial Code (12). This system has been continuously improved through the use of procedures such as risk-based surveillance and multi-criteria analysis (10,13). However, the magnitude of the epidemic may be dependent on factors associated with transmission dynamics and biosecurity standards (14) that have not been assessed. Since these factors vary from country to country, studies in different countries are warranted to support prioritization strategies.

Some studies indicate the benefits of modeling according to their conditions (15-18). Lewis *et al.* (15), based on HPAI simulation; recommend the most effective policy for HPAI control in their region of interest. Given Cuba's AI-free status, modeling could be useful for the improvement of the established disease surveillance system and decisions on resource allocation for disease prevention and poultry industry expansion. This study aimed to estimate the magnitude of the consequences of HPAIV incursion in Cuba and the effectiveness of important control measures.

MATERIALS AND METHODS

Study location and population dataset

The study examined Cuban commercial poultry facilities using data from the poultry farm registry of the National Center for Animal Health (CENASA) of the Ministry of Agriculture (MINAG). Five hundred and ten observations from poultry farms in several provinces were included (Fig. 1). Farms with a population less than 300 birds were excluded from the analysis, assuming that they were facilities intended for self-supply in certain sectors and, therefore, of lesser importance. Final sample size consisted of 467 observations.

The information was processed in the software NAADSM (North American Model of Infectious Disease Dissemination), version 3.3.2 (19). For each poultry farm, the following data were recorded: type of production, size, breeding sites, location, and infectious disease status (latent, subclinical, clinical or removed). The type of production was obtained from the purpose of each farm registered in a national poultry population database and it is detailed in Table 1.
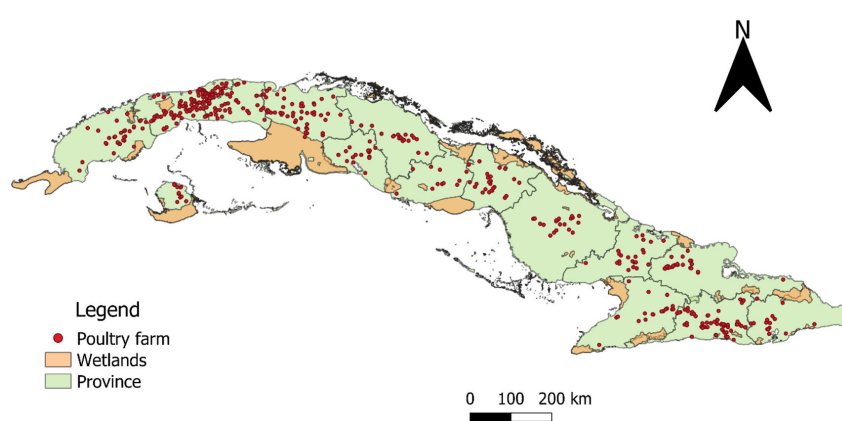

Figure 1. Distribution of commercial poultry farms assessed in the model. /
Distribución de las granjas avícolas comerciales evaluadas en el modelo.

Table 1. Production type categorization./ *Categorización de los tipos de producción.*

Purpose	Production type
Hens, broilers and their parents from zero to 45 days of age.	Starter
Hens, broilers and their parents after 45 days of age and before maturity.	Development
Layer hens, pheasant, layer breeders	Layer hens
Quails, layer quails	Quails
Turkey, turkey breeders	Turkey
Duck, ducks breeders, goose	Ducks
Broiler breeders, broilers	Broilers

Model parameters

Information from Lewis *et al*. (2015) and experts' opinion was used to represent the daily duration of HPAI (Table 2). Latency period was constant for all production types. BetaPERT distribution was used for subclinical and clinical infectious stages.

Spread

Simulations and direct and indirect contact were evaluated in different combinations depending on production type. For all cases, it was determined that farms could transmit subclinical disease. Frequency was considered as a variable, which was calculated using Poisson distribution, with data obtained from an experts' survey (19). Data obtained from this survey are shown in Table 3. Propagation of probabilities within a 1-km radius due to aerogenic transmission were considered according to Montserrat *et al*. (20). The area (in angles) exposed to risk and the time

it takes for the virus to spread by air were also surrounded. For this purpose, the Exponential Distribution with a moderate value of five days was selected.

Infection model

Three different levels of biosecurity were defined (95 %, Low; 50 % Medium and 10 %, High), to assess the probability of disease transmission. This information at NAADSM was the probability of infection transfer (if the source was positive) during probable contact between poultry.

Movement restrictions were categorized by two levels: [i] higher effectiveness and [ii] lower effectiveness. It was described through a linear function (x, y), according to the probability of virus transmission in time when movement was executed.

Surveillance processes were focused on the length of time that flocks remained infected. For this purpose, detection process was represented, which was influenced by the probability over time of observing

Table 2. Daily duration of HPAI for different period of production type. / *Duración diaria de la influenza aviar altamente patógena para los diferentes periodos de los tipos de producción.*

Production type	Latent	Subclinical Infectious	Clinical Infectious	Removed
Starter	0	Min(1),Max(16),Moda(6)	Min(2),Max(21),Moda(14)	270
Development	0	Min(1),Max(16),Moda(6)	Min(2),Max(21),Moda(14)	270
Layer hens	0	Min(1),Max(16),Moda(6)	Min(2),Max(21),Moda(14)	270
Quail	0	Min(1),Max(16),Moda(6)	Min(2),Max(21),Moda(14)	270
Turkeys	0	Min(1),Max(7),Moda(2)	Min(2),Max(21),Moda(7)	270
Ducks	0	Min(1),Max(21),Moda(7)	Min(2),Max(21),Moda(14)	270
Broilers	0	Min(1),Max(7),Moda(2)	Min(2),Max(21),Moda(7)	270

Table 3. Experts´ survey results on the frequency of direct and indirect contact between the different types of production. / *Resultados de las encuestas aplicadas a los expertos sobre la frecuencia de contacto directo e indirecto entre los diferentes tipos de producción.*

Combination of production types	Frequency of direct contact times/year				Media	Media/day
	Survey 1	Survey 2	Survey 3	Survey 4		
Starter- Development	3	3	3	5	3.5	0.009589041
Starter - Layer hens	0	0	0	0	0	0
Development - Layer hens	1	3	3	1	2	0.005479452
Layer hens - Layer hens	0	0	0	0	0	0
Ducks - Ducks	0	0	0	0	0	0
Ducks -All species	0	0	0	0	0	0
Turkeys - Turkeys	0	0	0	0	0	0
Quail - Quail	0	0	0	0	0	0
Quail - Turkeys	0	0	0	0	0	0
Contact type	**Frequency of indirect contact (times/year)**				**Media**	**Moda**
	Survey 1	Survey 2	Survey 3	Survey 4		
Feed supply	10	0	0	35	11.25	0
Technical views	24	25	15	15	19.75	15
Other visits (maintenance, admirative, etc.)	0	0	0	0	0	0
Vaccination or beak trimming	10	10	10	10	10	10
Egg collection	0	0	0	0	0	0
Total daily	0.4493					
Expected daily total	0.5479					
Poisson Distribucion	0.57816268					

clinical signs and the probability over time of reporting them once observed. Detection resulted from the interaction of those two probabilities. Accordingly, three detection speed levels were assessed: (i) rapid, equivalent to three days, (ii) moderate, corresponding to five days, and (iii) slow, equal to seven days.

Direct and indirect contact among farms was assessed days before detection for contact tracing and the probability of successful tracing and independent values for each type of production. Three risk zones were analyzed to represent the spread of the disease according to the distance to the infected farm: (i) High (3 km), (ii) Medium (5 km) and (iii) Low (10 km) according to WOAH (21,22). For each production type, it was evaluated whether or not detection on infected farms and tracing of direct and indirect contacts would create a limited outbreak. Poultry depopulation efficacy was assessed on the basis of the delay in days to carry out depopulation (1 day) and the capacity for this procedure. For this purpose, it was estimated how many days (x) and how many farms would be depopulated (y), by means of the relational function (x (1,2,3,7), y (1,1,2,5)).

Summarizing, 20 variables were analyzed through NAADSM: "contact frequency rate", "probability of transmission of infection", "movement restrictions", "delay in contact", "distance distribution", "probability of airborne transmission", "maximum distance of outbreak in km", "delay in virus spread", "probability of observing clinical signs", "probability of reporting clinical signs", "days before detection (direct and indirect)", "probability of tracing the event", "delay in tracing", and "previous delay to initiate this program". Depopulation capability, destruction priority, latency time, subclinical status time, clinical status time, and immune status time were also determined.

Simulated outbreak was assessed in at least one dormant farm and up to three simultaneously infected farms, depending on the number of dormant farms in the baseline. Therefore, three selection criteria were considered.:

1. Random selection through NAADSM, allowing inserting a number of farms in a dormant state.

2. Biased selection by population size (High, Medium, Low).

3. Selection by proximity to migratory waterfowl settlements as a risk factor and by proximity among farms within a radius of less than 5 km. Biased selection by population size (High, Medium, Low).

Simulated scenarios

One thousand iterations were completed in each simulation. Three replicates with 54 observations were analyzed for the scenarios according to index selection. Two hundred and sixteen scenarios, based on combinations of the three biosecurity levels, two movement restriction levels and three detection speed levels, were processed to represent different likely scenarios for AIV introduction.

Statistical analysis

Descriptive statistics were performed for the results of random selection, population size and risk scenarios. (i) The best scenario was: high biosecurity, higher animal movement restriction and detection of infection in 3 days. (ii) The most likely scenario was: higher probability of presentation according to Cuban characteristics and conditions, medium biosecurity and moderate detection speed (5 days). (iii) The worst scenario was: low biosecurity, less effective movement restriction and slow detection speed (7 days).

Statistical processing of the simulation results was run on R server (v. 3.5.0) and IDE (Integrated Development Environment) of RStudio development (v.1.1.447). Biosecurity level, movement restrictions and detection speed were identified as qualitative categorical independent variables. Dependent variables were infected farms, depopulated farms, slaughtered animals, spread duration, and disease duration. To evaluate the results, the Multivariate Analysis of Variance (MANOVA) was applied. The results in these scenarios were contrasted with confidence intervals with 95% confidence level.

A multiple linear regression model was applied indistinctly to discern the probability of association among the measures established to control the disease and the breeding sites that may become infected in an HPAI outbreak. A least squares test was performed on residuals to assess the goodness of fit of the model. For this analysis, the dependent variable was constructed as the percentage increase in the number of infected farms, $(V_f-V_i)/V_f$; V_f representing the number of farms infected at the end of the outbreak and V_i the number of farms initially infected (3 farms). Similarly, the probability of persistence of farms in a dormant state was assessed, using multiple logistic regression. Latent variable model of dichotomous data was constructed with values equal to one in positive cases, and equal to zero in negative cases.

RESULTS

It was found, in the best-case scenario by random selection, that the outbreak did not exceed the initially infected farms. Detection occurred within four days of infection and the first destruction within six days, with outbreak duration of 12 days. However, 87 826 dead animals were estimated, of which 37 888 were of the laying type.

In the most likely scenario, outbreak duration was 14 days, with an increase of one infected farm over the initial ones and an estimated loss of 199 913 poultry.

Epidemic curves showed that the highest number of infected animals was within 95th percentile.

The worst scenario involved 15 more farms than the best-case scenario, with persistent of farms in a latent state (Fig. 2), implying over a million of infected animals. On most farms, outbreak duration ranged from 20 to 26 days, almost two times longer than in the best scenario.

Statistical analyses of randomized scenarios, MANOVA, indicated that biosecurity and movement restriction were significant at the 99 % confidence level with respect to the dependent variables. (Annex 1). The interaction of biosecurity, movement restriction and detection speed had a 90 % confidence level with respect to the spread of HPAI (Annex 2). The least squares test for residuals revealed that biosecurity and movement restriction were significant at 95 % respectively. The model fitted the observed values with the predicted values, indicating a p-value of 0,0008804 (Annex 3). The fitted multiple linear regression model showed that low biosecurity and less movement restriction were the factors increasing infection with 99 % and 95 % confidence, respectively (Annex 4). Scenarios with low biosecurity had 57 % of the total positive cases.

For the high population size condition, the worst-case scenario, compared to the most favorable ones, implied a significant growth of the affected farms, highlighting the appearance of the latent state (Fig. 3).

Cases with a high population were tested and the detection rate for the worst case scenario was too slow at seven days (Fig. 4). It is evident that for rapid detection, infected and destroyed farms were not affected for the different biosafety levels. For the most likely scenario, the best scenario in terms of the number of depopulated and infected farms did not show significant differences.

Statistical analyses in high population size scenarios showed that biosecurity and movement restrictions were significant at 99 % and 90 % confidence level, respectively (Annex 5). Interactions between biosecurity and movement restrictions were significant at a 95 % confidence level (Annex 6). The least squares test showed that biosecurity and minor movement restriction had a 95 % confidence level (Annex 7). This model offered a goodness of fit of 0.7051(R2) according to the least squares test with a p-value = 0,00624. For the multiple binary regression, it was evident that movement restrictions was directly related with the increase of infected farms, which was significant at a 95 % confidence level (Annex 8).

There was a higher probability of dissemination on farm contiguity, making them priority regions for resilience development.

In the contiguous scenarios, the difference in losses between the worst-case scenario relative to the best-case scenario and the most likely scenario was
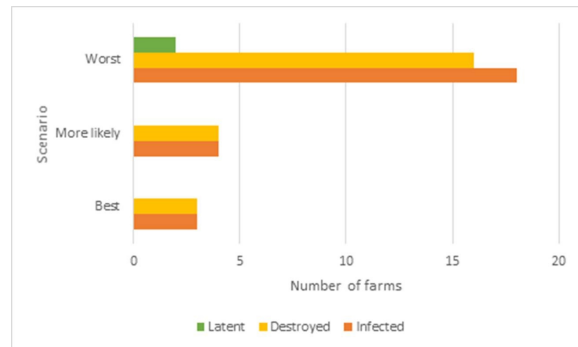

Figure 2. Expected farms affected by categories in different scenarios under random selection condition. / *Unidades afectadas estimadas, distribuidas por categorías en los escenarios de selección aleatoria.*
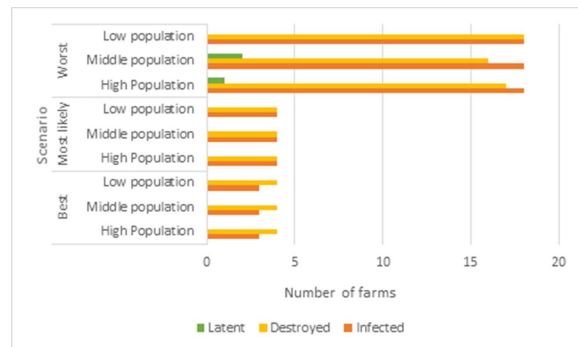

Figure 3. Expected farms affected by categories in different scenarios under population size condition. / *Unidades afectadas, esperadas divididas por categorías en los escenarios de densidad poblacional.*
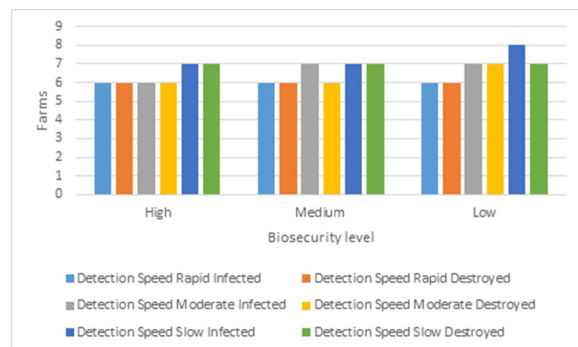

Figure 4. Expected number of categories of affected farms by detection time rate in high population size combined with biosecurity level and major movement restriction. / *Número previsto de categorías de unidades afectadas por tasa de tiempo de detección en tamaño de población alto combinado con nivel de bioseguridad y mayor restricción de movimientos.*

evident (Fig. 5). Even for the worst scenario, there were so many infected animals that farms persisted in a dormant state. With respect to animal losses, the best scenario had 198,178 infected animals and 218,076 slaughtered animals. Similar results were obtained in the most likely scenario; however, the worst scenario had almost four times as many infected animals as the rest.
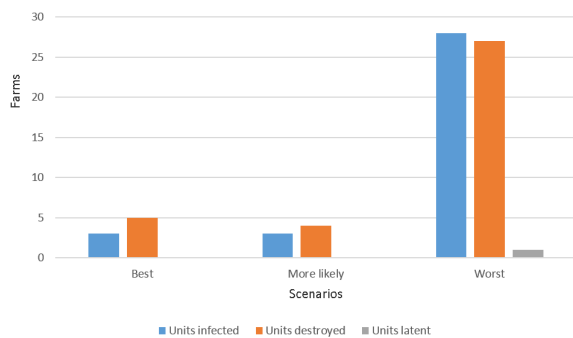
Figure 5. Expected farms affected by categories in different risk scenarios by farm contiguity . / *Unidades previstas afectadas por categorías en los escenarios riesgo por contigüidad de granjas.*

In scenarios close to migratory waterfowl settlements, the most likely had 2.65 times more slaughtered animals than the best scenario and it had almost two times less loses than the worst case. The outbreak for the worst scenario occurred in 27 days. In contrast, the other scenarios ranged from 10 to 12 days.

Comparison of the three main risk dimensions (high population size, farm contiguity and proximity to wetlands), for the worst-case scenario, showed the greatest impact for farm contiguity (Table 4).

Statistical analysis of farm contiguity revealed that biosecurity was significant with respect to infected farms at the 95 % confidence level and movement restrictions at the 90 % confidence level (Annex 9). Interactions between biosecurity and movement restrictions were significant for a 90 % confidence level with respect to infected farms (Annex 10). The least squares test showed that biosecurity was significant at a 95 % confidence level (Annex 11). This model showed a goodness of fit of 0,6389 (R^2) with a p-value = 0.01868. Regression model indicated that low biosecurity was significant at the 95 % confidence level with respect to the percentage increase in infection. These results were consistent with those obtained in previous scenarios showing the implication for virus spread caused by low biosecurity in the farms. (Annex 12).

DISCUSSION

The current study provided the first insight into potential cases for of HPAIVs transmission in Cuba through NAADSM. It showed referent values for containment measures for the successful management of eventual emergencies due to the incursion of these viruses in commercial poultry farms. The main objective of contingency plans for exotic infectious diseases is to restore a disease-free status as quickly as possible (23). This is the case of Cuba for several transboundary diseases, including HPAI; therefore modeling can be an important contribution in this regard (24).

The finding that biosecurity was one of the most influential factors in the impact of HPAI highlighted the importance of prevention. Biosecurity measures have been shown to be a major obstacle to the spread of HPAI on poultry farms (25,26). Indeed, prevention and preparedness are "peacetime" phases of disaster management and risk reduction (22,27). In particular, when facing infectious diseases with zoonotic/pandemic potential such as avian influenza, investing in prevention is not only cost-effective, but can also preserve human infections with their potential negative consequences (28,29). Moreover, as the main biosecurity measures are not disease-specific, the overall risk of infection with other infectious agents is reduced.

The finding that laying included the highest losses could be related to its higher contact rates compared to other types of production and to the movement of flocks from farm to farm from hatching to pullets. This movement rate, coupled with regular egg collection, could increase the likelihood of infection from other farms. In addition, the proportion of laying hens is the most represented commercial type in the whole country (ONEI 2021). This fact allows to insist on the importance of adequate biosecurity and enhanced surveillance. A recent study identified that the most significant farm-level risk factor for HPAI in table-egg commercial farms is being located within an existing control zone (30), while the supply chain is also identified as an important risk factor (31).

Although considering that all the breeding sites in Cuba may have similar probability of virus introduction (randomness), it would not be the most viable cost-effective surveillance (32). These scenarios helped to demonstrate that when facing the constant threat of HPAIV, the importance of reaching a high level of biosecurity to control or mitigate the risk of spread of these viruses should be emphasized. Similarly, Ssematimba *et al*. (22) report that poor biosecurity measures were the main reason for the continued spread of HPAI H7N7 in the Dutch poultry population, at a time when contingency plans had been put in place as a means to end the outbreak.

The lack of differences among affected farms according to population size highlights the importance

Table 4. Summary of losses in the three main risk conditions for the worst-case scenario. / *Resumen de pérdidas en los tres escenarios de riesgo para el peor escenario posible.*

Risk condition	Infected farms	Infected animals	Depopulated farms	Slaughtered animals
Higth Population size	18	1360430	17	1353030
Farm contiguity	28	785202	27	708303
Proximity to wetlands	10	279025	10	279025

of the interaction among risk factors, including biosecurity. In fact, Sharkey *et al.* (23) demonstrated the risk of infection in areas of high density produced on small and large scale, as well as Stevenson *et al.* (17) and Green *et al.* (35). However, the cases of low containment effectiveness demonstrated by simulation can be explained by the persistence of farms in a dormant state.

The present study revealed the need to detect infections less than three days after they have occurred to reach adequate mitigation. Currently, high-throughput diagnostic techniques providing results in less than 24 hours are widely available, even in Cuba (36, 37). Nevertheless, a study based on outbreak data (38) estimates that the most likely time of HPAI introduction ranges from ≤14.8 to ≤ 18.8 days before disease detection, depending on the poultry species. In fact, incubation period for HPAI infection is set at 14 days according to the Code (1).

To ensure a timely response, stakeholders' awareness is crucial to promote self-reporting of unusual mortality or the presence of signs or lesions related to HPAI infection. Detection of the pathogen may depend on human capacity and reaction to the manifestation of HPAI clinical signs. Maintaining and assessing production and mortality records, and reporting when abnormalities are observed according to syndromic surveillance procedures, also provides early warning and are mandatory in some situations (39,40). On the other hand, knowledge and training are among the main components to control HPAIVs (41).

Detection window is taken into account for contact tracing which, together with other important control measures such as movement restriction and stamping out, aims to prevent or reduce the size of an epidemic (42).

The model suggested that it is possible to contain the spread of HPAIVs for a detection within three days of infection, a first depopulation within six days and an outbreak duration of 12 days. Prompt identification of an infectious disease is crucial, but it is useful only if facilities can respond quickly and appropriately to the emergency situation in such a way that all necessary measures are implemented to contain and then progressively eliminate the infection (23).

Avian influenza virus control strategies in poultry vary depending on whether the target is prevention, management, or eradication (41) . Even within the same target, the severity of control measures may vary from country to country depending on the scope of contingency plans and the expected consequences of the disease. In fact, despite criticism, preventive stamping out of uninfected at-risk farms has been considered for several high-consequence diseases, including HPAI (16, 43-46).

Such decisions may depend on several factors including the magnitude and timing of the epidemic, as well as its implications for poultry and poultry product exports. Modeling is a useful resource for early decision making, particularly when facing of the potential incursion of pathogens causing high-consequence diseases. Poultry production in Cuba is predominantly based on laying, and constitutes one of the main sources of native animal proteins supporting an average per capita consumption of about 210 eggs/person/year (47). Hence it is an important component of food security (48).

The main limitation of this stochastic model was the need to establish, by different hypotheses, variables that could not be measured. Although the expert´s survey was useful to collect primary information, it could incur in the omission of significant data. In addition, in Cuba there have never been outbreaks of HPAIV infection in poultry. Therefore, there is no case available to compare the results and validate this study, considering that this is the first time that the presence of HPAIV has been simulated in Cuba.

Previous Cuban studies on this subject have strengthened surveillance system by stratifying the areas of greatest risk of disease occurrence (10,21,24) . These areas of interest, together with the results obtained in the present study, can provide early warning and resilience in case of outbreaks caused by HPAIVs.

The quick dissemination of infection in farm contiguity is consistent with the potential for aerogenic transmission attributed to the virus associated with the surprising magnitude of the HPAI epidemic in the United States during 2015 (50), which were taken into account. Therefore, areas with high farm contiguity should be prioritized for investment of resources in biosecurity and early warning of virus introduction. Decisions of this nature would reduce the risk of introducing the agent into poultry populations and even, in the event of its introduction, detect it quickly and initiate a timely response to contain the spread.

According to the results obtained in this study, special attention should be paid to improve biosecurity and to promptly impose movement restrictions after the detection of suspected or confirmed cases. Preventive stamping out was not taken into account in this model, although it has been used by several countries (45,51,52). However, there are large differences among countries in terms of the intensity of production systems and market aspects suggesting not to directly transfer this measure. In fact, a similar model in Canada showed no improvement in HPAI containment by this measure (16).

Strategic and systematic vaccination against HPAI was not taken into account in this model, even though it is a topic that is currently receiving much attention as an additional protective measure (53). However, this would require further studies, considering that vaccination is not a substitute for lack of biosecurity.

CONCLUSIONS

The study highlights the importance of strengthening preventive measures and effective control policies to limit or at least mitigate the spread of HPAIV under commercial poultry farming conditions in Cuba. Biosecurity proved to be the most influential factor requiring special attention in the prevention phase of emergency management and disaster risk reduction. This model suggests that it is possible to contain the spread of HPAI virus if detection is reached within three days of infection and depopulation of animals is completed within six days. This requires a high level of producer awareness promoting early warning and effective timely response.

REFERENCES

1. WOAH. Infection with high pathogenicity avian influenza viruses [Internet]. Chapter10.4 in Terrestrial Animal Health Code; 2022 [citado 21 de noviembre de 2022]. Disponible en: https://www.woah.org/en/what-we-do/standards/codes-and-manuals/terrestrial-code-online-access/

2. OIE. Report of the Meeting of the OIE Terrestrial Animal Health Standards Commission [Internet]. 2021. Disponible en: https://www.woah.org/fileadmin/Home/eng/Internationa_Standard_Setting/docs/pdf/A_TAHSC_Feb_2021_Report_Part_A.pdf

3. Sims LD, Domenech J, Benigno C, Kahn S, Kamata A, Lubroth J, et al. Origin and evolution of highly pathogenic H5N1 avian influenza in Asia. Veterinary Record. 2005;157(6):159-64.

4. Ramey AM, Hill NJ, DeLiberto TJ, Gibbs SE, Camille Hopkins M, Lang AS, et al. Highly pathogenic avian influenza is an emerging disease threat to wild birds in North America. The Journal of Wildlife Management. 2022;86(2):e22171.

5. Napp S, Majó N, Sánchez-Gónzalez R, Vergara-Alert J. Emergence and spread of highly pathogenic avian influenza A (H5N8) in Europe in 2016-2017. Transboundary and Emerging Diseases. 2018;65(5):1217-26.

6. Swayne DE, Sims L, Brown I, Harder T, Stegeman A, Delgado M, et al. Strategic challenges in the global control of High Pathogenicity Avian Influenza [Internet]. Paris: World Organisation for Animal Health; 2023. (90th General Session). Disponible en: https://www.woah.org/app/uploads/2023/05/a-90sg-8.pdf

7. Shi J, Zeng X, Cui P, Yan C, Chen H. Alarming situation of emerging H5 and H7 avian influenza and effective control strategies. Emerging microbes & infections. 2023;12(1):2155072.

8. Lycett SJ, Pohlmann A, Staubach C, Caliendo V, Woolhouse M, Beer M, et al. Genesis and spread of multiple reassortants during the 2016/2017 H5 avian influenza epidemic in Eurasia. Proceedings of the National Academy of Sciences. 2020;117(34):20814-25.

9. Aguilar S, Manica LT, Acosta M, Castro R, Hernández Z, González A, et al. Spatio-temporal patterns of waterbird assemblages in Cuba's south coast wetlands: conservation implications. Wetlands. 2019;1-13.

10. de las Nieves Montano D, Irian Percedo M, Vioel Rodríguez S, Fonseca O, Centelles Y, Ley O, Abreu Y, Delgado B, Capdevila Y, Regis Santoro K, Quesada T. Influenza aviar. Oportunidades de mejora del sistema de vigilancia activa basado en riesgo en Cuba. Revista de Salud Animal. 2020 Dec;42(3):1-10.

11. WAHIS. Cuba - Influenza de tipo A de alta patogenicidad (Inf. por los virus de la) (aves que no sean de corral, incluyendo las silvestres) (2017-) [Internet]. Cuba; 2023 [citado 27 de noviembre de 2023]. Report No.: 10. Disponible en: https://wahis.woah.org/#/in-review/4895?fromPage=event-dashboard-url

12. WOAH. Animal health surveillance [Internet]. Chapter 1.4. in Terrestrial Animal Health Code; 2022. Disponible en: https://www.woah.org/en/what-we-do/standards/codes-and-manuals/terrestrial-code-online-access/?id=169&L=1&htmfile=chapitre_surveillance_general.htm

13. Ferrer E, Alfonso P, Ippoliti C, Abeledo M, Calistri P, Blanco P, et al. Development of an active risk-based surveillance strategy for avian influenza in Cuba. Preventive Veterinary Medicine. 1 de septiembre de 2014;116(1):161-7.

14. Do O, Ca M, Ab A, Ai A, Ma L, Fo K. Re-emergence of Highly Pathogenic Avian Influenza H5N1 in Nigeria, 2014-2016: Role of Social Network and Value Chain Forces in Interstate Transmission. Virologica Sinica [Internet]. agosto de 2020 [citado 8 de junio de 2021];35(4). Disponible en: https://pubmed.ncbi.nlm.nih.gov/32236814/

15. Flores U, Rivera A, Sanchez J. Uso del Modelo Norte Americano de Dispersión de Enfermedades - NAADSM, para la Simulación de Brotes de Fiebre Aftosa en el Perú, en Base al Movimiento de Bovinos - Perulactea [Internet]. 2014 [citado 24 de abril de 2018]. Disponible en: http://www.perulactea.com/2014/05/01/uso-del-modelo-norte-americano-de-dispersion-de-enfermedades-naadsm-para-la-simulacion-de-brotes-de-fiebre-aftosa-en-el-peru-en-base-al-movimiento-de-bovinos/

16. Lewis N, Dorjee S, Dubé C, VanLeeuwen J, Sanchez J. Assessment of Effectiveness of Control Strategies Against Simulated Outbreaks of Highly Pathogenic Avian Influenza in

Ontario, Canada. Transbound Emerg Dis. junio de 2015;64(3):938-50.

17. Pepin KM, Spackman E, Brown JD, Pabilonia KL, Garber LP, Weaver JT, et al. Using quantitative disease dynamics as a tool for guiding response to avian influenza in poultry in the United States of America. Preventive veterinary medicine. 2014;113(4):376-97.

18. Stevenson MA, Sanson RL, Stern MW, O'Leary BD, Sujau M, Moles-Benfell N, et al. InterSpread Plus: a spatial and stochastic simulation model of disease in animal populations. Preventive veterinary medicine. 2013;109(1-2):10-24.

19. Prieto AF. Distribuciones de probabilidad. Cundinamarca, Colombia: Universidad de Cundinamarca, Facultad de Ingenieria, Programa de Ingenieria Industrial,Sede Soacha; 2015 p. 21.

20. Torremorell M, Alonso C, Davies PR, Raynor PC, Patnayak D, Torchetti M, et al. Investigation into the airborne dissemination of H5N2 highly pathogenic avian influenza virus during the 2015 spring outbreaks in the Midwestern United States. Avian Diseases. 2016;60(3):637-43.

21. Mongkolsawat C, Kamchai T. GIS Modeling for Avian Influenza Risk Areas | IJG:International Journal of Geoinformatics. International Journal of Geoinformatics [Internet]. 2009 [citado 7 de enero de 2024];5(1); ISSN. 1686-6576. Disponible en: http://creativecity.gscc.osaka-cu.ac.jp/IJG/article/view/483

22. Health (OIE) WO for A. Guidelines on Disaster Management and Risk Reduction in Relation to Animal Health and Welfare and Veterinary Public Health. OIE Paris, France; 2016.

23. Dalla Pozza M, Ceolin C, Marangon S. Emergency response following suspicion of an avian influenza outbreak. Zoonoses and public health. 2008;55(1):50-3.

24. Clemmons EA, Alfson KJ, Dutton III JW. Transboundary animal diseases, an overview of 17 diseases with potential for global spread and serious consequences. Animals. 2021;11(7): 2039. https://doi.org/10.3390/ani11072039

25. Authority (EFSA) EFS. Animal health and welfare aspects of avian influenza and the risk of its introduction into the EU poultry holdings - Scientific opinion of the Panel on Animal Health and Welfare. EFSA Journal. 2008;6(6):715.

26. So H, Bae Y, Mo I. Risk assessment of highly pathogenic avian influenza by comparison of biosecurity level in domestic poultry farms. Korean Journal of Poultry Science. 2019;46(4): 313-25.

27. Bonbon E, Myers L. Good emergency management practices: a review of the core principles, with a focus on preparedness. Revue Scientifique et Technique (International Office of Epizootics). 2020;39(2):533-41.

28. Bernstein AS, Ando AW, Loch-Temzelides T, Vale MM, Li BV, Li H, et al. The costs and benefits of primary prevention of zoonotic pandemics. Science Advances. 2022;8(5):eabl 4183.

29. Gilbert W, Adamson D, Donachie D, Hamilton K, Rushton J, 2023. A Cost-Benefit Analysis of Preparing National Veterinary Services for Transboundary Animal Disease Emergencies. Transboundary and Emerging Diseases, 2023. 1765243

30. Green AL, Branan M, Fields VL, Patyk K, Kolar SK, Beam A, et al. Investigation of risk factors for introduction of highly pathogenic avian influenza H5N1 virus onto table egg farms in the United States, 2022: a case-control study. Frontiers in Veterinary Science. 2023;10, 1229008: 1-14. https://doi.org/10.3389/fvets.2023.1229008.

31. Moyen N, Hoque MA, Mahmud R, Hasan M, Sarkar S, Biswas PK, et al. Avian influenza transmission risk along live poultry trading networks in Bangladesh. Scientific reports. 2021; 11(1):19962.

32. Alfonso P, Miranda EF, Al E. Perfeccionamiento del sistema de vigilancia activa de la influenza aviar basado en el análisis de riesgo de su introducción al país. Anales de la Academia de Ciencias de Cuba [Internet]. 23 de diciembre de 2016 [citado 21 de mayo de 2018];6(3). Disponible en: http://www.revistaccuba.cu/index.php/acc/article/view/495

33. Ssematimba A, Hagenaars TJ, de Wit JJ, Ruiterkamp F, Fabri TH, Stegeman JA, et al. Avian influenza transmission risks: analysis of biosecurity measures and contact structure in Dutch poultry farming. Preventive veterinary medicine. 2013;109(1-2):106-15.

34. Sharkey KJ, Bowers RG, Morgan KL, Robinson SE, Christley RM. Epidemiological consequences of an incursion of highly pathogenic H5N1 avian influenza into the British poultry flock. Proceedings of the Royal Society B: Biological Sciences. 2008;275(1630):19-28.

35. Green AL, Branan M, Fields VL, Patyk K, Kolar SK, Beam A, et al. Investigation of risk factors for introduction of highly pathogenic avian influenza H5N1 virus onto table egg farms in the United States, 2022: a case-control study. Frontiers in Veterinary Science. 2023;10. https://doi.org/10.3389/fvets.2023.1229008

36. WOAH. Terrestrial Manual. Chapter 3.3.4. Avian Influenza (Infection with Avian Influenza Viruses [Internet]. 2021. Disponible en: https://www.woah.org/fileadmin/Home/eng/Health_standards/tahm/3.03.04_AI.pdf

37. Alfonso P, González CL, Becerra HC. Animal influenza in the Caribbean: a systematic review of research and surveillance. Revista de Salud Animal. 2020;42(3): 1- 15.

38. Hobbelen PH, Elbers AR, Werkman M, Koch G, Velkers FC, Stegeman A, et al. Estimating the introduction time of highly pathogenic avian influenza into poultry flocks. Scientific Reports. 2020;10(1):12388.

39. Hernández-Jover M, Phiri BJ, Stringer L, Martínez Avilés M. Developments in animal health surveillance. Front. Vet. Sci. 7:637364. https://doi.org/10.3389/fvets.2020.637364

40. Gonzales JL, Hobbelen PH, van der Spek AN, Vries EP, Elbers AR. Highly pathogenic avian influenza A H5 virus outbreaks in broiler farms in the Netherlands. Clinical signs, transmission and identification of reporting thresholds. bioRxiv. 2023;2023.01. 05.522008.

41. Sims LD, Swayne DE. Avian influenza control strategies. Chapter Animal influenza. Animal influenza. 2016;363-77.

42. Klinkenberg D, Fraser C, Heesterbeek H. The effectiveness of contact tracing in emerging epidemics. PloS one. 2006;1(1):e12.

43. Bergevoet RHM, van Asseldonk M. Análisis económico de la erradicación de brotes de fiebre aftosa usando estrategias alternativas de control. Archivos de medicina veterinaria. 2014;46(3): 381-8.

44. Boender GJ, Hengel R van den, Roermund HJ van, Hagenaars TJ. The influence of between-farm distance and farm size on the spread of classical swine fever during the 1997-1998 epidemic in The Netherlands. PLoS One. 2014; 9(4): e95278.

45. Salvador R, Tanquilut N, Macmac R, Na Lampang K, Chaisowwong W, Pfeiffer D, et al. Evaluation of strategies using simulation model to control a potential outbreak of highly pathogenic avian influenza among poultry farms in Central Luzon, Philippines. PloS one. 2020; 15(9):e0238815.

46. Bradhurst R, Garner G, Roche S, Iglesias R, Kung N, Robinson B, et al. 2021. Modelling the spread and control of African swine fever in domestic and feral pigs. Technical Report. Australian Government, Department of Agriculture, Water and the Environment. Disponible en: https://era.daf.qld.gov.au/id/eprint/8363/

47. Oficina Nacional de Estadística e Información, Sitio en Actualización [Internet]. 2020 [citado 13 de septiembre de 2022]. ONEI. Disponible en: http://www.onei.gob.cu/

48. Bradhurst R, Garner G, Hóvári M, de la Puente M, Mintiens K, Yadav S, et al. Development of a transboundary model of livestock disease in Europe. Transboundary and Emerging Diseases. 2022;69(4):1963-82.

49. Hernández S, Zamora A. Análisis geoespacial para la apreciación del riesgo de influenza aviar en Cuba. En La habana, Cuba; 2019. Disponible en: http://www.informaticahabana.cu/en/node/4396

50. Zhao Y, Richardson B, Takle E, Chai L, Schmitt D, Xin H. Airborne transmission may have played a role in the spread of 2015 highly pathogenic avian influenza outbreaks in the United States. Scientific reports. 2019;9(1):11 755.

51. Andronico A, Courcoul A, Bronner A, Scoizec A, Lebouquin-Leneveu S, Guinat C, et al. Highly pathogenic avian influenza H5N8 in south-west France 2016-2017: A modeling study of control strategies. Epidemics. 2019;28:100340.

52. Seymour RG, Kypraios T, O'Neill PD, Hagenaars TJ. A Bayesian nonparametric analysis of the 2003 outbreak of highly pathogenic avian influenza in the Netherlands. Journal of the Royal Statistical Society Series C: Applied Statistics. 2021;70(5):1323-43.

53. Alqazlan N, Astill J, Raj S, Sharif S. Strategies for enhancing immunity against avian influenza virus in chickens: A review. Avian Pathology. 2022;51(3):211-35.

ANNEXS

Annex 1. MANOVA in random scenarios

	Df	Pillai	Approx. F	Num Df	Den DF	Pr(>F)
Biosecurity	2	0.93174	6.8323	12	94	1.021e-08 ***
Residuals	51					

Movement	1	0.8812	71.209	5	48	2.2e-16 ***
Residuals	52					

Annex 2. Results of interaction between independent variables in random scenarios

	Df	Pillai	Approx. F	Num Df	Den DF	Pr(>F)
Biosecurity: Movement:Velocity	17	1.3149	1.6524	51	108	0.01516 *
Residuals	36					

Annex 3. Least squares test to the residuals in random scenarios

	Df	Sum Sq	Mean Sq	F Value	Pr(>F)
Movement	1	0.36682	0.36682	7.8475	0.007315**
Biosecurity	2	0.65855	0.32928	7.0443	0.002077 **
Speed	2	0.14907	0.074453	1.5945	0.213575
Residuals	48	2.24370	0.04674		

Annex 4. Multiple linear regression model in random scenarios

Deviance Residuals:				
Min	**1Q**	**Median**	**3Q**	**Max**
-0.47413	-0.12949	-0.00566	0.14227	0.49223
Coefficients:				
	Estimate	**Std.Error**	**T value**	**Pr(>|t|)**
(Intercept)	0.2077	0.7207	2.883	0.005879 **
SpeedModerate	-0.01195	0.7207	-0.166	0.869020
SpeedFast	-0.11695	0.7207	-1.623	0.111195
MovementLess	0.16484	0.05884	2.801	0.007315**
BiosecurityLow	0.26636	0.07207	3.696	0.000562***
BiosecurityMedium	0.17402	0.07207	2.415	0.19611*

Annex 5. MANOVA in high population size scenarios

	Df	Pillai	Approx. F	Num Df	Den DF	Pr(>F)
Biosecurity	2	1.1986	4.8609	8	26	0.0009569 ***
Residuals	15					

Movement	1	0.5833	4.5494	4	13	0.01619*
Residuals	52					

Annex 6. Results of interaction between independent variables in high population size scenarios

	Df	Pillai	Approx. F	Num Df	Den DF	Pr(>F)
Biosecurity: Movement	5	2.2031	2.9424	20	48	0.001138**
Residuals	12					

Annex 7. Least squares test to the residuals in high population size scenarios

	Df	Sum Sq	Mean Sq	F Value	Pr(>F)
Movement	1	0.27415	0.274152	9.7491	0.008816**
Biosecurity	2	0.42220	0.211102	7.5070	0.007683 **
Speed	2	0.11030	0.055150	1.9612	0.183251
Residuals	12	0.33745	0.028121		

Annex 8. Multiple linear regression model in high population size scenarios

Deviance Residuals:				
Min	**1Q**	**Median**	**3Q**	**Max**
-146377	-42166	5467	45551	239222
Coefficients:				
	Estimate	**Std.Error**	**T value**	**Pr(>|t|)**
(Intercept)	996546.5	70435.4	14.148	7.57e-09 ***
SpeedModerate	505.5	70435.4	0.007	0.99439
SpeedFast	-23243.0	70435.4	-0.330	0.74710
MovementLess	213632.3	57510.2	3.715	0.00296**
BiosecurityLow	42597.2	70435.4	0.605	0.55660
BiosecurityMedium	17497.8	70435.4	0.248	0.80801

Annex 9. MANOVA in contiguous farms scenarios

	Df	Sum Sq	Mean Sq	F Value	Pr(>F)
Movement	1	26.889	26.889	5.9024	0.02727 *
Residuals	16	72.889	4.5556		
Biosecurity	2	183.44	91.722	6.4796	0.00937**
Residuals	15	212.33	14.156		

Annex 10. Results of interaction between independent variables in contiguous farms scenarios

	Df	Sum Sq	Mean Sq	F Value	Pr(>F)
Biosecurity:Movement	5	254.44	50.889	4.3208	0.0176*
Residuals	12	141.33	11.778		

Annex 11. Least squares test to the residuals in contiguous farms scenarios

	Df	Sum Sq	Mean Sq	F Value	Pr(>F)
Speed	2	58.914	29.457	2.0885	0.166609
Movement	1	21.040	21.040	1.4917	0.245400
Biosecurity	2	219.516	109.758	7.7819	0.006809**
Residuals	12	169.252	14.104		

Annex 12. Multiple linear regression model in contiguous farms scenarios

Deviance Residuals:				
Min	**1Q**	**Median**	**3Q**	**Max**
-0.50000	-0.15278	0.02778	0.16667	0.66667
Coefficients:				
	Estimate	**Std.Error**	**T value**	**Pr(>|t|)**
(Intercept)	-1.667e-01	2.152e-01	-0.775	0.45357
SpeedModerate	3.333e-01	2.152e-01	1.549	0.14729
SpeedFast	4.540e-17	2.152e-01	0.000	1.00000
MovementLess	1.111e-01	1.757e-01	0.632	0.53895
BiosecurityLow	6.667e-01	2.152e-01	3.098	0.00922**
BiosecurityMedium	1.667e-01	2.152e-01	0.775	0.43357