

eISSN:2703-1322

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#### **Research** article

# DiFLUsion: A novel space-time alert system for HPAI

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Article History: Received: 1-Sept-2024 Accepted: 28-Sept-2024 \*Corresponding author: Iglesias Irene iglesias@inia.csic.es

#### Abstract

The emergence and spread of highly pathogenic avian influenza (HPAI) pose significant threats to global poultry industries, wildlife, and public health. Migratory waterfowl play a critical role in its propagation. The current epidemic of HPAI since late 2020 is unprecedented due to a severe increase in incidence, a constant presence without seasonality, greater susceptibility in wild birds, more frequent jumps to wild and domestic mammals (including cats, cows, and mink), and spread to new regions, presenting new challenges in disease control. These changes in the dynamics of the disease have resulted in numerous outbreaks and the death of millions of domestic and wild birds, directly impacting poultry product costs and representing a persistent threat to the poultry industry and public health. In response to this challenge, we present DiFLUsion, an innovative early warning system for HPAI that has been operational in Spain since 2021. DiFLUsion enables early detection in disease-free zones connected by wild bird movements to HPAI-affected areas. This system integrates multiple data sources and analytical tools using Python and ArcGIS, allowing for the modulation of alerts based on the location of HPAI outbreaks in Europe, the seasonality of wild bird movements, and virus survival temperatures. DiFLUsion operates in real-time, updating weekly, sending alerts to the Spanish Ministry of Agriculture, and providing an interactive map viewer. DiFLUsion has effectively identified high-risk areas for HPAI introduction in Spain, aligning with recent outbreak patterns. Over three years, the system generated 77,872 alerts, demonstrating a sensitivity of 85.43% for high-risk categories and a specificity of 74.48% for the highest alert level. These alerts have facilitated preemptive measures, enhancing response times and mitigation strategies. In conclusion, DiFLUsion enhances the decision-making capacity of livestock health managers in Spain, enabling them to prepare for and respond proactively to avian influenza epidemics.

Keywords: Avian influenza, Wild birds, Surveillance, Real-time alert system

**Citation:** Albáñez-Porras, P., de la Torre, A., Gómez-Pérez, J. I., Tomás-Tenllado, C., García, E., Cáceres, G., Pérez, A., Iglesias, I. 2024. DiFLUsion: A novel space-time alert system for IAAP. Ger. J. Vet. Res. 4 (4): 31-42. https://doi.org/10. 51585/gjvr.2024.4.0107

#### Introduction

Highly pathogenic avian influenza (HPAI) represents a global threat due to the devastating economic losses for the poultry industry, the impact on wildlife, and its recognized zoonotic potential. Economic losses occur in the poultry industry not only as a result of its high transmissibility and mortality rates but also due to closed borders and trade restrictions in affected areas. High mortality also poses a threat to critically endangered wild bird species (Gamarra-Toledo et al., 2023; Sacristán et al., 2024). HPAI also poses a threat to humans because they can become infected with HPAI subtypes such as H5N1, H7N9, and H9N2 (WHO, 2023). Since 2020, an unprecedented global epidemic of HPAI has been occurring. The HPAI H5N1 clade 2.3.4.4b virus caused the death of over 311 million domestic birds and thousands of wild birds worldwide between 2021 and 2024 (WOAH, 2024) The current HPAI H5N1 subtype has shown changes in its epidemiological dynamics, with increased incidence, constant presence without seasonality, greater susceptibility in wild birds, and more frequent jumps to mammals (Sacristán et al., 2024). These changes have led to the infection of new areas, such as South America, and infections in various mammal species, posing a threat to biodiversity conservation (Adlhoch and Baldinelli, 2023).

Additionally, domestic animals like minks (Agüero et al., 2023; Kareinen et al., 2024) and cats (reported in Poland, South Korea, and the USA), as well as cows, have been infected. Transmission has been identified between minks (Agüero et al., 2023) and between cows (Caserta et al., 2024), representing a direct threat not only to production but also to public health. Therefore, a better understanding of avian influenza is required from a One Health perspective to ensure safe poultry production, minimize the economic consequences of an introduction, optimize prevention strategies such as real-time alert systems and biosecurity of holdings, avoid risks of zoonotic avian influenza viruses in terms of public health, and protect wild bird populations in terms of conservation and ecology.

Wild birds are the natural reservoir of avian influenza virus (AIV) (Hill et al., 2012), especially aquatic wild birds (Martelli et al., 2023). The movements of migratory waterfowl play a crucial role in the intercontinental and continental spread of HPAI (Caliendo et al., 2022; Lo et al., 2022), and transmission at the wildlife-domestic interface is recurrent despite biosecurity and control measures. Contact with waterfowl was identified as the main risk factor for the primary introduction of avian influenza into poultry during holdings several epidemic waves worldwide (Gonzales et al., 2017; Ramos et al., 2017).

In Spain, the risk of HPAI introduction by wild birds from affected countries is significant, especially during the autumn migration of water birds (Martinez et al., 2009). These introductions are inevitable; however, real-time monitoring of the disease and wild bird movements from affected areas enables anticipation and targeted reinforcement of surveillance and control measures in the poultry industry based on current risk levels. This approach is supported bv recommendations from the World Organization for Animal Health, WOAH (WOAH, 2023), and the Commission Delegated Regulation (EU) 2020/687 of Dec 17, 2019 (EC, 2020). The Commission's decision advises

member states to ensure increased surveillance of wild bird populations and to regularly review and adjust measures to maintain risk mitigation and reinforce biosecurity, adapting to the prevailing epidemiological situation, particularly concerning the risks posed by wild birds.

To address this, DiFLUsion, an automated real-time alert system, was developed in Spain in 2021 (Iglesias et al., 2022) to identify weekly risk areas for the introduction of HPAI through wild bird movements. After three years of use with real data, the purpose of this study was to demonstrate and evaluate this newly developed avian influenza alert system (DiFLUsion) in Spain as a tool for analyzing the risk factors associated with the introduction of HPAI into Spain via migratory wild bird movements by monitoring and visualizing the risk weekly. Both descriptive and analytical procedures were included in this analysis. This alert system operates in real-time and is updated weekly, sending alerts to the end user (the Ministry) weekly. An interactive web viewer with scroll-time features for outbreaks and alerts is included in the DiFLUsion package, along with information on migratory wild bird movement data. This surveillance system has the potential to enhance the design of existing national surveillance plans and serve as a valuable tool for avian influenza surveillance and decision support.

# Material and methods

## Data sources

The alert system was developed based on the premises explained in the introduction: i) Wild birds serve as natural reservoirs of AIV, and ii) Migratory movements of wild birds play a critical role in the spread of the AIV. Accordingly, the model captures the potential spread of the disease through wild bird movements from regions reporting HPAI outbreaks to regions that have not yet been affected. To establish these connections, data on wild birds' movements, specifically from ringing and recovery efforts provided by the Spanish Society of Ornithology (SEO/Birdlife), were employed. Using ringing and recovery data of wild birds from the Spanish Society of Ornithology (SEO), The temporal dynamics of these movements were constructed using bird observation data collected in citizen science databases. As a result, the model integrates various input databases for the modeling of migratory routes and the calculation of risk values:

## **Risk bird species**

The aquatic species at highest risk for the introduction of HPAI into Spain through migratory movements from affected areas have been identified. This selection includes 139 wild bird species considered high-risk based on specific criteria: i) Identified as high risk for Spain (Martinez et al., 2009) for H5N1 HPAI; ii) Highlighted by the Commission Decision of Jun 25, 2010 (EC, 2010) and European Food Safety Agency, EFSA (Gonzales et al., 2017), evaluated considering Spain's specific ornithological situation in terms of species representativeness (census and records available from SEO/Birdlife); and iii) Inclusion of species affected by recurrent outbreaks not listed in previous assessments based on WOAH outbreak notifications since 2002-2024 (WAHIS, 2023).

## Wild bird migratory movements

We consider migratory movements as the set ringing-recovery data of wild birds provided by SEO/Birdlife. The records include information on the ringed species, ringing and recovery coordinates, ringing/recovery dates, and other fields that are not included in the model. In total, 66,764 ringing and recovery records corresponding to the 139 selected wild bird risk species were included. To streamline the data for model use, the following definitions are established: i) Ringing Point: The coordinates of a ringing event located outside Spanish territory. ii) Collection Point: The coordinates of a recovery event located within Spanish territory.

Note that both definitions are complementary within the same record: if a coordinate of a route is considered as ringing, the remaining coordinate must be a collection point, and vice versa.

#### **Probability of movement**

To estimate this probability, we used the phenology data for each species provided by SEO ornithological experts. This data is based on their expert knowledge and the e-Bird citizen science databases (eBird, 2023) on bird sightings. The combination of both data sets allows us to determine the weekly variation in the presence of each bird species in Spain, which is assumed to represent the potential probability of movement.

## Forecasted minimum temperature

The survival of the HPAI virus in the environment is inversely related to temperature,

with lower temperatures being critical for the virus's persistence (Brown et al., 2009; Songserm et al., 2006). The relationship between the virus's survival time and ambient temperature is modeled using a regression formula (Iglesias et al., 2011), as presented in equation (2).

To calculate the maximum weekly survival time, which is associated with the highest risk value, the weekly average minimum temperature for each considered region is used, with data obtained from AEMET (State Meteorological Agency) through OpenDATA, a REST API generated by the agency.

## **HPAI** outbreaks

The data was obtained weekly from WOAH-WAHIS (WAHIS, 2023). The DiFLUsion algorithm is designed to be compatible with any other database available in CSV format, allowing for the incorporation of additional or alternative data if needed. From 2020 through July 2024, the database comprised 17,972 HPAI notification records in Europe, including both outbreaks in domestic and wild birds, a number that continues to increase each week. Since the risk movements of wild birds relevant to Spain originate from Europe (Martinez et al., 2009), only outbreaks located on the European continent are selected, and outbreaks reported within the study country (Spain) are excluded, as the model's function is to alert the introduction of the disease.

#### Administrative unit

For Spain, the livestock region has been selected as the primary administrative unit. This choice is based on its widespread use by veterinary authorities, for whom the tool is specifically designed for HPAI surveillance management in the country. The livestock region represents an administrative unit of intermediate size between a municipality (Local Administrative Unit, LAU, according to European legislation) and a province (NUTS3 level from European legislation) (EC, 2003). Spain is divided into a total of 496 livestock regions.

## Model algorithm

The system operates by monitoring HPAI notifications for both domestic and wild birds in Europe, as reported by WOAH, and then checking around the location of each notification for the presence of records of wild bird movements to Spain (ringing points). If such records are found, alerts are generated at their corresponding collection points in Spain, with the risk and alerts

modulated based on species and temperature factors. The complete model is described as follows:

Time-period: For a given week, "s" HPAI notifications are selected through a 12-week (3-month) time window for the week for which the risk is to be assessed, i.e. [s - 12, s), under the assumption that no outbreaks have occurred in the week being predicted.

of Movements wild birds HPAI and notifications: Around each new HPAI in Europe, a 25-kilometer buffer (representing the mean daily migration distance of the 25 species at highest risk for HPAI infection (Iglesias et al., 2010). Ringing points located within the buffer area are considered, and the related "collection point" in Spain is activated, together forming the potential routes of HPAI introduction in Spain. It should be noted that a route may be activated due to multiple outbreaks in Europe, which means that the situation of greatest risk is prioritized, and a route can be activated multiple times as necessary depending on how many times it falls within different influence areas.

Alerts: The activated routes are transformed into alerts, with a risk value R assigned to each livestock región G in Spain (496 in total) calculated as follows:

$$R_{G} = \left(\sum_{i}^{r} \varepsilon_{i} \cdot p_{mov,i}\right) \cdot t_{sup,g}$$
(1)

where  $\varepsilon_i$  represents the type of outbreak (domestic, wild, or captive, assigned respective weights of 1, 0.5, and 0.1) that has activated route *i* with collection in livestock region *G*,  $p_{mov,i}$  represents the probability that the ringed and collected species through route *i* are found in Spanish territory and  $t_{sup,g}$  is the virus survival time.

Virus survival time (days) is dependent on temperature and is calculated as follows:

$$t_{sup,G} = -7.82 \cdot \ln(T_G) + 29.94 \tag{2}$$

whereas  $T_G$  is the minimum temperature value recorded for livestock region *G*, considered through the mean value of a spline interpolation of the minimum temperature predicted by AEMET for the week under consideration.

To smooth the risk value R across each of the 496 G livestock regions and their neighboring regions in Spain, an interpolation method is applied to the entire Spanish territory, divided into 5 areas: peninsular Spain, the Canary Islands, the Balearic Islands, Ceuta, and Melilla. The interpolation method used is the Inverse Distance Weighting (IDW), using ArcGIS PRO software (version 3.2, ESRI Inc.; Redlands, CA, USA).

Finally, since the primary objective in developing the alert system was to produce userfriendly alerts for decision-making, detailed weekly reports and data presentation on an interactive map were included. GIS technology is used to visually display alerts through a web which application, provides retrospective information on alerts and outbreaks from previous periods. Additionally, an automatic weekly email is sent to veterinary authority officials or other interested personnel, containing the information displayed as a map in image format and a detailed report in spreadsheet format.

The model has been programmed in Python 3.9, using various libraries and packages, such as the Esri-licensed package *arcpy*, which allows for geospatial data processing. Code in R has also been used to obtain the validation results by utilizing the 'pROC' library (Robin et al., 2011). This R code is executed from the main Python code to automate the comprehensive analysis of the tool.

# Results

The following are the results of the retrospective evaluation of alerts generated by the DiFLUsion system for the period from 01/01/2021 to 31/12/2023, during which a total of 77,872 alerts were produced. During this period, 355 HPAI notifications were reported in Spain (253 in wild birds, 94 in poultry farms, and 8 in captive animals). The model's design allows for the extraction of information on potential HPAI introduction pathways in the study area, facilitating an analysis of the countries, migratory routes, and wild bird species that generated the highest number of alerts and their associated risk values, as well as identifying the regions in Spain with the highest risk.

## Countries contributing to alerts

During this period, alerts were predominantly generated by migratory routes originating from 34 different countries. Germany was the leading contributor to alerts, responsible for the highest number of activated routes and associated risks. The Netherlands and France were followed by Switzerland and the Netherlands in terms of the number of alerts and the risk value (Table 1). specific countries in the epidemiological These findings underscore the importance of landscape of HPAI in Spain.

Country	Total risk (%)	Total count routes (%)
Germany	37.3	31.01
Switzerland	16.28	10.82
Netherlands	15.61	17.04
United Kingdom	9.83	11.02
Belgium	8.89	8.55
France	6.79	12.06
Sweden	1.49	1.63
Denmark	1.32	2.02
Poland	0.72	1.84
Hungary	0.27	1.29

**Table 1:** Top 10 countries generating the highest number of alerts. The "Total risk (%)" column shows the percentage of the total risk value associated with each country, while the "Total count routes (%)" column indicates the percentage of routes relative to the total of 17,289,568 routes that generated alerts.

#### Species contributing to alerts

A total of 132 species have generated alerts through more than 17 million activated routes. The common white stork (*Ciconia Ciconia*) is identified as the species generating the most alerts and the highest risk, followed by the lesser black-backed gull (*Larus fuscus*) and the blackheaded gull (*Larus ridibundus*) (Table 2). The dominance of these species highlights their critical role in the transmission dynamics of HPAI from affected areas to Spain.

**Table 2:** Top 10 wild bird species generating the highest number of alerts and associated risk. The "Total risk (%)" column indicates the percentage of the total risk value associated with each species, while the "Total count Routes (%)" column shows the percentage of routes relative to the total of 17,289,568 routes that generated alerts.

Species	Total risk (%)	Total count routes (%)
Ciconia ciconia	55.6	45.1
Larus fuscus	21.06	18.92
Larus ridibundus	6.26	7.56
Vanellus vanellus	3.73	4.3
Phalacrocorax carbo	1.41	2.27
Anser anser	1.09	1.93
Ardea cinerea	0.85	1.42
Larus melanocephalus	0.84	1.86
Platalea leucorodia	0.57	1.23
Sterna hirundo	0.36	1.29

# High-risk areas in Spain: identification and spatial and temporal distribution

The DiFLUsion system, as an output of the model, assigns risk values on a weekly basis to each of the 496 livestock regions in Spain, allowing for the identification of areas with elevated risk. There is a notable concentration of regions on high alert in the southern (Huelva, Cádiz), northeastern (Catalonia), and central (Madrid) parts of the peninsula and around the

coast livestock regions.

The risk detected by the DiFLUsion system shows a clear seasonal pattern, with the highest risk values occurring in January and February. There is a noticeable decline in risk values from March through July, with the lowest value in July. This decline could correspond to reduced migratory wild bird activity and an increase in temperatures (conditions less favorable for virus persistence). Risk values increase again at the end of the year, in November and December, suggesting a renewed risk likely associated with

the return of migratory wild birds and seasonal environmental factors.



**Figure 1:** Map of Spanish livestock regions on alert level 5 (2021-2023) with reported influenza outbreaks during this period. The color gradient represents the number of weeks each region sustained this alert level during the specified period, with the scale ranging from yellow (1-40 weeks) to dark red (121-157 weeks). Points represent reported HPAI outbreaks within the same timeframe, categorized as captive (white), domestic (black), or wild (green). In the bottom right, three Spanish territories distant from the peninsula and assessed separately are shown: Ceuta, Melilla, and the Canary Islands (from left to right).



**Figure 2:** Total risk values of highly pathogenic avian influenza (HPAI) in Spain, aggregated monthly during the period 2021-2023. The graph illustrates the fluctuations in HPAI risk throughout the year, highlighting seasonal peaks and trends in risk distribution over the three years.

## Model validation

To evaluate the predictive capability of the model, sensitivity and specificity analysis were conducted using the ROC (Receiver Operating Characteristic) curve and the AUC (Area Under the Curve) evaluation. For the period from 01/01/2021 to 31/12/2023, the execution of the model generated a total of 77,872 alerts. Each alert was assigned a positive or negative boolean value depending on whether the livestock region had reported an outbreak within 8 weeks (considering the two weeks before, the same week, and the three weeks after), increasing the number of alerts with a positive case to 899. A matrix was contingency then generated, including true positives, true negatives, false positives, and false negatives, and sensitivity and specificity were calculated based on their definitions.

ROC curve analysis identified the optimal risk thresholds for categorizing alerts into different levels, ranging from 0 to 5. The curve, which illustrates the relationship between sensitivity and specificity for different decision thresholds, and AUC is calculated to quantify the overall performance of the model. Figure 3 shows the ROC curve and the associated AUC value of 0.82, indicating that the model has an 82.31% probability of distinguishing between positive and negative cases. To determine the optimal threshold of risk values, the option of the shortest distance to the ideal point on the ROC curve is used, which allows defining from which risk value a prediction is considered positive, minimizing the possibility of false positives and false negatives. Based on this threshold, six alert

categories (from alert 0 to alert 5) are established to weigh the risks, adjusting sensitivity and specificity according to the category. The category graph in Figure 4 shows the distribution of alerts in positive cases according to the alert category, highlighting the percentage of positives reported in alert 5 (63.29% of the alerts generated with a risk value associated with this category correspond to a positive case). Summing the percentages of alerts 3, 4, and 5, the total number of positive cases covered by these categories reaches 85.43%.

Finally, the validation results for alert categories 3, 4, and 5 are summarized, providing comprehensive view of the model's ิล performance. Table 3 shows the threshold value that determines the separation between categories, as well as the sensitivity and specificity values associated with them. The highest sensitivity corresponds to alert 3, with a percentage of 85.43%, meaning that 17 out of 20 alerts generated with a risk value of 200 or higher are truly positive cases. For specificity, alert 5 reflects the highest percentage, with 74.48%. This implies that approximately one-fourth of the alerts with a risk value of 600 or higher were considered positive when they were actually negative, leading to greater precision. The balance between both extreme categories is maintained by alert 4, with a sensitivity of 78.42%, meaning that more than three-quarters of the alerts generated with a risk value of 300 or higher correspond to positive cases, and a 64.15%, specificity of resulting in an overestimation error of positive cases close to one-third.

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Level risk	Threshold	Sensitivity %	Specificity %
0	1	100	3.36
1	100	93.99	45.39
2	150	89.10	52.40
3	200	85.43	57.26
4	300	78.42	64.15
5	600	63.29	74.48



**Figure 3:** ROC curve analysis for the DiFLUsion model during the period 01/2020 – 12/2023. The area under the ROC curve (AUC) value indicates a correct classification rate of 82.31%.



Distribución de alertas en diagnósticos positivos

**Figure 4:** Graph representing the percentage of true positive cases detected by the model from January 2020 to December 2023, organized by alert categories (A0 to A5). Each category shows the corresponding percentage of detection, with the total number of Highly Pathogenic Avian Influenza (HPAI) outbreaks noted in parentheses.

## Discussion

The implementation and evaluation of DiFLUsion, a novel real-time alert system for HPAI, underscore its effectiveness in predicting and managing the risk of HPAI introduction into Spain. The system demonstrates a high degree of accuracy, with a sensitivity of 85.43% for highrisk alerts and a specificity of 74.48% at the highest alert level. These metrics are further reinforced by an Area Under the Curve (AUC) value of 0.82, which indicates a strong ability to differentiate between true positive and negative cases. These findings provide compelling evidence that DiFLUsion can effectively identify areas at risk, offering timely alerts that are for risk-based surveillance crucial and preventive measures. This is particularly significant given the dynamic nature of HPAI and its potential to cause widespread outbreaks.

DiFLUsion's ability to adjust to real-time data and generate alerts based on evolving conditions is a key feature that aligns it with other predictive models used for HPAI and other infectious diseases (Kjaer et al., 2023; Gargallo et al., 2022; Bhatia et al., 2021). This dynamic adaptability not only enhances ongoing surveillance efforts but also supports proactive risk management strategies. A distinguishing advantage of DiFLUsion is its incorporation of a wide range of wild bird species that are particularly relevant to Spain, identified as potential vectors for HPAI introduction. Many of these species were initially recognized during the 2006 HPAI epidemic by Martinez et al. (2009), and the current model has expanded this list through collaboration with ornithological experts from SEO BirdLife. This targeted approach ensures that the model is finely tuned to the ecological and epidemiological context of Spain, thereby improving its predictive accuracy and operational utility. In contrast to more generalized models like EFSA's Bird Flu Radar (Gargallo et al., 2022), which provide valuable insights across Europe through a homogeneous framework, DiFLUsion offers a more tailored solution for the specific Spanish context (For instance, of the 12 species included in the Bird Flu Radar model at the time of this study, only 4 are representatives for the Spanish context) by focusing on the most relevant avian species and migratory patterns in Spain.

DiFLUsion's real-time capabilities offer significant advantages for HPAI surveillance and prevention strategies in Spain by facilitating the

generation of weekly alerts and providing detailed spatial analyses of risk areas. This functionality enables the targeted reinforcement of surveillance and biosecurity measures, ensuring that interventions are both timely and effective. The results align with previous experience in managing avian influenza in Spain, where the southwest and northeast regions, including some of the country's most significant wetlands, have consistently been identified as high-risk zones for wild bird surveillance (Martinez et al., 2009). These wetlands, such as Doñana and the Ebro Delta, serve as critical wintering habitats for numerous species of aquatic wild birds, underscoring the strategic importance of these regions in HPAI surveillance and monitoring efforts (De Juana and Garcia, 2015; Claudino-Sales, 2019). Moreover, the seasonal pattern of HPAI risk, as captured by DiFLUsion, clearly reflects its cyclic nature, with pronounced peaks occurring during the winter months in the Northern Hemisphere. The highest risk levels are observed from January to March, followed by a notable resurgence in late autumn (November and December). However, the key added value of DiFLUsion lies in its ability to provide risk assessments on a weekly basis and at the level of specific livestock regions. For instance. traditionally high-risk regions such as the southwest of Spain (Andalucía) and the northeast (Catalonia) were identified as high-risk areas by the model.

Additionally, regions like Madrid (in the center), which are not typically considered highrisk, also experienced a high concentration of outbreaks. This validates the model's predictions and highlights its utility in informing public health and agricultural policies. While winter remains the most critical period for HPAI risk, ongoing changes in the dynamics of the disease (Sacristan et al., 2024) have led to out-of-season occurrences, which DiFLUsion has effectively identified, such as along the eastern coast of Spain during the summer of 2023. In essence, DiFLUsion enables focused surveillance on a week-by-week basis and facilitates the timely identification of emerging high-risk zones. This allows authorities to implement preventive actions, such as confining outdoor poultry farms during periods of elevated risk. Consequently, the system has been actively used by the Spanish Ministry of Agriculture, Fisheries, and Food since

surveillance and control efforts.

The results from the DiFLUsion model highlight Germany's significant role as a source of HPAI alerts, which aligns with the extensive spread and impact of HPAI observed in the country, particularly along the North Sea coast and within the Wadden Sea region. Recent studies, such as Pohlmann et al. (2024), have documented substantial HPAI outbreaks in Germany, notably during the summer of 2022. Additionally, the high number of alerts generated by DiFLUsion in relation to Germany can be attributed to other factors, such as that Germany is situated along key migratory routes that connect various regions of Europe, including those leading to Spain, making it a critical junction for the transmission of HPAI via migratory birds.

Despite its strengths, DiFLUsion has certain limitations that need to be addressed in future iterations. One significant limitation is the reliance on wild bird ringing and observation data, which may not capture the full complexity of migratory patterns and HPAI transmission dynamics. There may be species-associated biases, as larger species are more easily observed, not only when detecting weak or dead wild birds due to HPAI in passive surveillance but also because they are more frequently ringed and recovered, given their visibility at a distance. The results showed that the common white stork (Ciconia ciconia) generated the most alerts, followed by the lesser black-backed gull (Larus fuscus) and the black-headed gull (Larus ridibundus). Although the stork is a large species, it is also one of the most significant migratory birds in Spain during the winter (Molina and Del Moral, 2005). In addition to the migratory populations, Spain hosts expanding resident populations that interact with these wintering individuals.

Similarly, while black-headed and lesser black-backed gulls are primarily migratory, Spain supports growing also resident populations that mingle with the wintering individuals 2009). (Molina, Continuous outbreaks of HPAI in black-headed gulls across Europe during April and May of 2022 maintained a high-risk level in the DiFLUsion system for Spain in June (WAHIS, 2023). This was followed by numerous notifications in July and August along the entire Mediterranean coast, involving

2021, serving as a crucial tool in HPAI black-headed gulls and other species with similar habitats, such as terns (WAHIS, 2023). These patterns suggest that the outbreaks in Spain may have resulted from introductions from these affected European populations.

> Additionally, the model's dependence on temperature data for virus survival introduces variability that may not fully reflect real-world conditions. This is similar to challenges faced in other models, such as those discussed by Kjær et al. (2023) and Ward et al. (2008), where environmental factors like landscape and distance to water bodies played significant roles in HPAI transmission but were difficult to quantify precisely.

> Another limitation is the potential for both overreporting and underreporting of HPAI outbreaks, which can impact the accuracy of the DiFLUsion model's alerts. For instance, Germany's detailed system of reporting, particularly the practice of documenting wildlife cases individually, significantly contributes to the higher alert counts generated by the DiFLUsion model. While these reporting practices provide valuable and granular data, they may introduce a reporting bias that inflates the number of alerts compared to regions with different, potentially less detailed, reporting standards.

> Conversely, the underreporting of outbreaks poses a different challenge, potentially leading to an underestimation of risk in certain areas. The endemic component of HPAI, particularly during 2022 in wild birds across Europe (EFSA, 2022), suggests that there may be more HPAI activity than is captured by current surveillance efforts. This under-reporting is especially concerning in areas with dense foliage or higher temperatures, where detecting carcasses is more challenging. Similar to other models facing these issues (Kjær et al., 2023), DiFLUsion could benefit from incorporating more comprehensive data sources, including those related to wild bird density, species behavior, and environmental conditions, to enhance its accuracy and predictive power. Despite these challenges, in the DiFLUsion system, notifications serve as a critical indicator of disease presence in a region, effectively triggering all necessary risk assessments and responses for Spain.

> The scalability and adaptability of DiFLUsion to other regions or countries are promising areas for future research. Expanding the model to

include additional data layers, such as specific wild bird migratory routes and climate change impacts on migration patterns, could improve its utility across different geographical contexts. Additionally, integrating DiFLUsion with other surveillance systems could provide a more comprehensive approach to managing HPAI globally.

Further studies should also explore the potential for DiFLUsion to differentiate between different HPAI strains and subtypes, as these may have varying transmission dynamics and risks. By refining the model to account for these differences, it may be possible to improve its predictive accuracy and the effectiveness of targeted interventions. The introduction of phylogenetic data to connect outbreaks would be highly useful, although it is very challenging to analyze all cases and conduct these analyses in wild birds.

## Conclusions

DiFLUsion represents a significant advancement in HPAI surveillance and risk management. Its real-time alert capabilities and spatial analysis offer valuable tools for decision-makers in Spain, enabling proactive responses to emerging threats. The model's successful validation and alignment with actual outbreak data suggest that it can play a critical role in enhancing national and regional disease control strategies. However, ongoing refinement and expansion of the model are necessary to address its current limitations and to fully realize its potential in protecting public health and the poultry industry from the threat of HPAI.

#### **Article Information**

- Funding. This research was funded by the CON20-171 (Spain), OMB-N°0925d-0001(USA), AEG21-198 (Spain), and HE (EU267457-EUPAHW).
- Conflicts of Interest. The authors declare no conflict of interest.
- Acknowledgments. We would like to thank the Spanish Ornithological Society for their expert ornithological advice and for sharing bird-ringing data.
- Authors contribution. P.I.: Data curation, Conceptualization, Formal analysis, Investigation, Software, Validation, Visualization, Writing – original draft. J.I. G.P. and C.T.T.: Formal analysis, Methodology, Software Supervision; CB and C.J. and E.V.: Software, Visualization; E.G. and G.C.: Funding acquisition and Supervision; AP and AT: Funding acquisition, Investigation. I.I.: Data curation, Funding acquisition, Conceptualization, Formal analysis, Investigation, Resources, Software, Validation, Visualization, Writing – original draft.

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