Graphical Abstract

Spatio-temporal analysis of A/H5N1 outbreaks in cats in Poland combining the evidence from official data, citizens reports and media interest

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Highlights

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- the A/H5N1 was already probably circulating in cats in second half of May in regions bordering with Ukraine and this information could be missing without extending surveillance to non official sources, so some other approaches without epiginetic-spatio-temproral analytics of multuple sources may lead to wrong epidemiological conclusions
- most of positive cases form chains structure both on bird migration paths and close to high abundance of nesting sites of water birds, thus We recommend to perform active monitoring (PCR and serology) of water birds in Pomerania and selected sites in Western Poland (as maybe some low viral pressure of active virus is still present at the end of July 2023).

Spatio-temporal analysis of A/H5N1 outbreaks in cats in Poland combining the evidence from official data, citizens reports and media interest

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Abstract

One Health topics such as transmission of zoonotic agents of domestic/wild birds and environmental residue problems, food and feed safety become increasingly important to modern society in the post pandemic times. We performed spatio-temporal analysis of A/H5N1 epizootic in cats in Poland in Spring-Summer 2023 based on: 1) 30 (positive) and 27 (negative) cases from WOAH reference lab, 2) suspected 87 cases submitted by animal owners (participatory epidemiology), 3) daily time series of i) Google queries for Avian Influenza (AI), cats disease and cats deaths, as well as ii) mentions of cat/cats and AI in social and traditional media; 19 RNA sequences of viruses; Data triangulation suggests the most likely scenarios based on Google queries, news and social media analysis, reference lab results and participatory epidemiology reports: 1) The first A/H5N1 cases were probably in cats in late May near the south-eastern border (based on the evidence from the retrospective media analysis and participatory reports). 2) The highest disease burden was in Pomerania (particularly Gdansk) in mid-June. 3) The highest positivity ratio was in Western Poland, indicating outbreak clusters in late June/early July. 4) Positive cases formed clear chains, supporting the environmental source hypotheses (on the connection to bird migration paths and nearness of nesting sites of water birds). 5) There are at least 2 (eastern and western Poland) separate genetic clusters of viruses. We recommend active serological/PCR monitoring in both water birds and environment in Gdańsk and some sites in Greater Poland area (probably there is still small viral pressure as the cases were in August). Even that hypothesis of poultry meat

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(being feed for cats) contamination, seems to be unlikely (i.e. our method indicated that positive cases situated far from bird migratory paths were wrongly geocoded), aviary disease surveillance system needs to be updated to handle with new pandemic threats.

Keywords: infectious disease modelling, epigenetics, spatio-temporal analysis, avian influenza, zoonotic disease of poultry

1. Introduction

Avian influenza (AI), particlulary highly pathogenic (HPAI) subtypes A/H5 is deadly (depending on bird species) and contagious disease of poultry and free living birds, being subject to mandatory eradication in European Union (EU). HPAI pose a serious threat to poultry industry and due to zoonotic/spillovers potential [1] also to other species (including humans), therefore the risk of virus (re-) emergence in environment and farms should be minimised. The main route of introducing the virus into Poland since 2020 is seasonal migration of wild birds [2]. Here, we report on the detection of spillover of A/H5N1 to cats during spring/summer 2023 in Poland. Isolated viruses from cats after 18.06.2023 in Poland belong to clade 2.3.4.4b and cluster with virus strains from birds sampled in Central Europe from late 2022 onwards [3, 4] and were found in wild birds thought spring/summer 2023 in Poland. Between May (26.05 Ketrzyn: Warmian–Masurian) and July (01.07 Kraków: Lower Poland) there were no outbreaks of AI in domestic poultry in 2023 (these mentioned are from different strains).

1.1. cats and birds in Poland

From last few years of observation the common risk areas of AI in wild birds include usually eastern and western parts of the country, and in particular, areas located within the territory of Greater Poland, Pomerania and Kuyavian-Pomeranian from the west and Subcarpatian, Lublin and Warmian–Masurian from the east [2]. By comparing locations of AI outbreaks in wild birds and poultry farms with risk factors (i.e. density of waterfowls) a significant spatial correlation was recorded [5]. Terrestrial and aquatic wild birds are the usually the primary source of outbreaks in domestic poultry, however clustering patterns demonstrate that secondary spread also played a role in high-density hubs of production in Greater Poland and Masovia regions [6]. The total population of on farms (2M) and domestic cats (2M) in Poland is about 4 million [7, 8] which shows that less than every third household in Poland has a cat [9]. This means that the density of cats per number of inhabitants is about 1.5-2 times lower than the European average [8]. Cats may be expose to the AI viruses directly by interactions with birds (i.e. hunting), or indirectly by contact with contaminated environment (i.e. formits or feed brought by humans to home).

1.2. related works on current outbreak

Rabalski et al. [3] involved a participatory approach with animal owners self-report and included in phylogenetic analysis isolates obtained from not-referenced labs collected without sanitary/veterinary supervision (which without understanding how the concept of infodemics could lead to wrong conclusions about distribution and cause of the infections). On the other hand, Domańska et al. [4] used the reference laboratory positive cases as the material (which without spatio-temporal context and non-including negative results could lead to missing important clustering and path forming patterns). Both approaches are complementary, but a kind of integrative approach is missing to incorporate strong and weak sides of each dataset. The authors [4, 3] adopted a number of simplifications, which should be justified, as papers may reach a wide audience and may be cited by one health community or journalists. Thus, no attempt has been made up to submission of this article to carry out a quantitative spatio-temporal-genetic assessment of the risk of virus introduction. Added benefits of the multi-source space-timeepigenetic vs only space, only time, only genetic modeling of epidemiological data (current state of the art) has been recently raised in epidemiological community [10].

1.3. research aims

Objective of comparative analysis and interpretation of data to determine:

- Triangulated distribution of diseases in time and space with genetic inference
- Presence or absence of disease
- source detection

2. Data and methods

The results are presented using:

- phylogenetic matrices with epidemiological inference;
- descriptive and exploratory analyses (as partially performed by [3, 4]
- time series approach as auto/cross-correlations and calculation of the epidemic curve characteristics. Temporal dynamics and Spatio-temporal clusters can explain disease transmission occurring through local and long-distance spread [11].
- a risk (and relative risk) map, clustering (k-mean and dbscan [12]) developed using Geographic Information System (GIS);
- spatio-temporal distribution of queries for cats health and AI related topics. Digital epidemiology approach is widely used, for example, "infoveillance" as a term comes from information surveillance [13].
- epigenetic-spatio-temporal matrices and clustering [10].

All analyses (expect some media interest spatio-temporal transformation with media listing tools as EventRgistry.org and Brand24.com [14] are fully reproducible as well as all data and recipes (with much more extended analysis) are available [15, 16, 17].

2.1. Participatory sample

Some activists started community-based surveillance/ participatory epidemiology [14] in social media (a phenomenon popular in Poland since COVID-19 pandemic [18, 19]). unofficial data - suspected 87 case submitted by animal owners. Here, we take the date of symptoms developing into account (from 15.06 to 08.07).

2.2. WOAH reference lab

According international regulations [20] on infectious disease control (particularly HPAI), the National Veterinary Research Institute in Puławy is responsible for testing materials from animals Thus we process official data (from reference lab) in an area of Poland (testing date from 22.06 to 10.07). It consists of 30 cases (positive results) and 27 controls (negative results). Here, we take the date of laboratory test into account (extended discussion on the choice and its consequences is available [16])

2.3. Infodemiology

Variable "GT interest [21]" is daily time series mean across AI, cats disease and cats deaths queries. Google Trends monitoring (on both national and voievodship level) can be extremely beneficial for tracking trends, understanding public demand of information.

Variable "media interest" is daily time series sum of mentions cat/cats and AI, cats in social and traditional media. Media listening involves tracking mentions of specific topics in the media (here related both to AI and cats health) and show that is the supply of information (mainly local news portals and agencies [22]). This can help to stay informed about the latest public perceptions, and potential crises. Brand24 was used for that goal.

Both tools allow for a real time listening [23] and collect information before the official surveillance started or after participatory epidemiology lost attention in reporting (for full time span 15.05-12.07).

3. Results

Until about 18.06 [14] we have a local discussion in Gdańsk (at least there are traces of it on Facebook) and interest in the environment of veterinarians [Fig. 1]. On June 19, the case gained momentum of a nationwide dimension (specialists and local Pomeranian media publicized the situation).

Since the disclosure of information about the alleged contamination of poultry meat with the A / H5N1 virus on June 29 [24], at a high level, the Veterinary Inspection's activities have finally started in full swing, because it affects the poultry industry, which is very important for the Polish economy, and "central control" has been turned on (certainly good from the information point of view), but perhaps not epidemiologically.

Due to the infodemic risks, the flow of information in the context of the H5N1 avian influenza situation was monitored by state and poultry organizations [24, 25]. Thus, certain stakeholders of the system could introduce mal/mis-information [26].

Based on all sources separately (all of which have their advantages and disadvantages) quite different conclusions can be stated, so an integrative framework is missing. For instance Rabalski et al., [3] did not attempt to differentiate signal from noise in citizens' databases [25, 27, 23, 15] Interpolated daily dynamics of positive case, suspected cases from participatory study, normalized interest in Google trends and No articles/post on the issue per million citizen in media suggest that the epizootic and infodemic have



Figure 1: Time evolution of the epizootic and infodemic

already passed the peak phase and is in the ending phase [fig. 1]. Variables "googles and media interest" are derivatives across AI, cats disease and cats deaths queries. However, upon submission of the article, there are still some individual positive case, which mean that the outbreak is not over.

3.1. Spatial

We see that all cats materials (together +/-, but mainly negative) and participatory reports are sampled moreover from general cats population (according to human population density) with bias towards city cats [fig. 2].

From a first look is seems to be in line with the water bird migration and nesting areas at the water reservoir routes [17]:

- north-south line along the Nysa Kłodzka, Oder, Warta and Vistula rivers;
- north-east line of the Vistula and Bug basin.





Figure 2: Geographical distribution of + and negative - laboratory cases of cats or ill suspected from participatory results

Under-representation of positive clusters in the most populated metropolies in Poland (Upper-Silesia agglomeration, Warsaw, Krakow, Łodź and Szczecin at least in reference lab data) suggests that human travels (with or without cats) doesn't play a role in the epidemic spread.

The spatially-varying probability of different type of point (positive and negative) using kernel smoothing suggest that in western Poland positivity ratio in Western Poland is a few folds higher than in rest of the country [16].

The highest social interest was observed in Pomerania and secondary in Kuyavian-Pomeranian and Lower Silesia [fig. 3]. Moreover, the most common geographical places till so called risk assessment phase [fig. 1] mentioned in the media were Gdańsk, Lublin, Poznań, Bygdoszcz [14].

We use Average Nearest Neighbour (ANN) first and second order (the average distance between all points within a dataset and their individual nearest or second point) to detect topological structure.

Positive data points have smaller ANN than suspected (even there is



Figure 3: Normalized (by population size) affection of regions across Polish voivodships

much less of them) and than negative results, thus it suggests that positive cases form some characteristics chains [tab. 1]. This support migratory bird cause of outbreak.

We see that social interest is strictly spatially correlated with participatory engagement [17].

3.2. Temporal

Lagged-correlation analysis suggests that citizens reports are one week ahead the positive cases. Interest in Google trends and in media is one week after positive cases [17].

The shape of the pseudo-epidemic curve of participatory reports is not symmetric [fig. 4]. One of the possible non-biological explanation is that people lost their attention in the beginning of July (Since July 8, no new cases were reported). The peak of submission reports (by symptoms developments) was around 14-17.06.

| Data | 1NN | 2NN |
|-----------------|------|------|
| Positive cases | 0.18 | 0.30 |
| Negative cases | 0.38 | 0.65 |
| Citizen reports | 0.20 | 0.36 |

| Table | 1. |
|-------|----|
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Among the fist portion of samples analyzed by WOAH reference lab 80% were positive. The positivity ratio went down to 20% till the end of interested period [16] and the deceasing trend is ongoing upon submission of this article.

By analysis of smoothed epidemic curve of positive results [fig. 4] and cumulative distribution [16], the peak of outbreak (by testing time) was around 27-30.06. The left arm of epidemic curve is missing. Possible biological explanation is that due to nesting time and need to feed offsprings, the wild birds penetrate bigger areas (bigger exposure for cats and human mechanical vectors). The non-biological possible explanation is that surveillance started when the outbreak was already after the peak.

Temporal correlations between series showed that there is de-synchronization between participatory reporting and supply/demand information needs.

3.3. Epigenetic

19 samples from WOAH reference [4] lab (from cats only) proved that there are at least 2 separate sources of infections (one in Western Poland and second dispersed around the country).

3.4. Spatio- Temporal

There is small bit consistent gradient between positive cases and participatory results between time and geography from East to West (-0.20 for reports+; -0.21 for citizens) and from South to North (0.23 for reports+; 0.16 for citizens), but not significant [17]. Temporal correlations between series on voivodeship level showed that there is de-synchronization between participatory reporting and supply/demand information needs [17].

There is clear spatio-temporal cluster of suspected cases in second part of Mai on Lublin/Podkarpacie area [fig. 7]. Time gives no additional information (in terms of quality of clustering) in reference lab positive cases.

In the in statu nastendi analysis [16] only the case of the Caracal of Lodz did not fit (wrongly coded place by veterinary inspection) and this analysis



Smoothed positive cases and suspeceted (from citizens) intensity in

Figure 4: Smoothed (pseudo)-epidemic curves

allowed to find this error. For negative cases, the Silesian cluster is mainly FIV/FELV and the Mazovian cluster is driven by other diseases causing symptoms (mainly neurological) imitating AI.

Spatial Clusters of negative cases are more integrated and hierarchical than for positives ones [fig. 7, tab. 2], which may be artefact of catching clusters of infectious diseases with similar symptoms. This definitely must be taken into account in interpretation of participatory reports [3]. High be-



Figure 5: Hierarchical matrices of SNPs distances: among 19 samples from WOAH reference lab [4]; 11 samples with environmental one [3]

Samples 1 "A.environment.Poland.Kra1.2023" and 11 A.cat.Poland.Kra1.2023." from [3] are from suspected cat case and its feed (poultry meat), which never been confirmed by WOAH reference lab. There are no clear separate clusters for the small sample from [3], which can lead to wrong conclusions (as authors didn't have big enough sample to see potential different sources). The cat meat case from the Kraków area is clustered with the Gdańsk subcluster. Moreover, Lublin region cases (from eastern Poland) mixes well with cases from Western Poland, which also made and illusion of disperse outbreak.

In the in statu nastendi analysis [17] we speculated, that virus found in meat in Krakow area (meat bought in beginning of June: "A.environment.Poland.Kra1.2023") should be close to (if our hypothesis of introduction thought Lublin/border area is valid) early cases from Lublin and difference in SNPs is less than 4.

| Data | betweeness | sum of squares |
|-----------------|------------|----------------|
| Positive cases | 5.49 | 6.14 |
| Negative cases | 3.71 | 4.84 |
| Citizen reports | 6.49 | 9.67 |

Table 2: K-mean statistics in XYT normalized model



Figure 6: K-means clustering

tweenees and Sum of squares of spatio-temporal clustering of positive cases [tab. 2] in comparison to negative cases and participatory reports (after taking into account different size of sample) suggests positive points are ordered in some kind of chains (i.e. corridors for wild birds movement).

3.5. Epigenetic - Spatio- Temporal

We propose combine toEuclidean distance for xY (space) and T (time) in normalized unites with additional layer of SNPs counts distance of viruses for 19 cases for each full information is available.

We see that a single Poznan case (upper left corner [fig. 7]) is from a separated introduction of virus. One big blocking cluster of Western Poland can be seen (with a one linking case from a Lublin region close to subcluster of cases from Gdańsk). However, links between Gdańsk nmetropolitan area and other regions maybe explained by being touristic destination hot-spot in the summer [28].

Weakly connected cluster of Lublin cases is complementing the hierarchical matrix.

According to DBSCAN there are 3 clear clusters (sensitive to choice of weights in inference model [fig. 8]). First is a single case in Poznan, second western Poland and third Lublin area. Second and third clusters are interlinked. We see strong sensitivity to the choice of the weight of different layers (spatial, temporal and genetic) or fixed number of clusters [17]..



Figure 7: Hierarchical matrix of epigenetic-spatio-temporal distances among 19 samples

4. Conclusions

If one can take into account possible biased of each method then, data triangulation suggest that most likely (according to the Google and media analysis, reference lab results and participatory epidemiology):

• the first cases A/H5N1 were probably in cats in second half of May in regions bordering with Ukraine (possible meat and wild life sources).



Figure 8: Dbscan clusters of epigenetic-spatio-temporal distances among 19 samples

This information could be missing without retrospective media analysis and participatory reporting;

- the biggest burden of disease was in Pomerania (in particular in Gdansk) in the middle of June;
- positivity ratio is the highest in Western Poland as well as western Poland form a sepereate epigenetic-spatio-temporal cluster, which suggest that the last outbreak clusters were in end June/beginning of July in north-Western Poland;
- the positive cases are clearly forming chains (in contradiction to negative cases and to participatory reports) which support environmental source hypothesizes;
- poultry meat contamination as additional factor of spread would explain some additional transmissions (i.e. caracal case in Lodź or cannot be easily explained by environmental exposure and this case was found to be wrongly geocoded or possible case in Cracow need deeper investigation).
- Trajectory of main events [fig. 1] in outbreak investigation (since mid May till mid July and still individual cases reported till submission of the article) suggest environmental long-term exposure.

4.1. Water bird migration and nesting

We suggest water birds serological passive sampling in Lubelskie province (probably there is no more viral pressure as the peak of cases was probably in May).

The hypothesis to be verified is that the most of positive cases form chains both on bird migration paths and close to high abundance of nesting sites of water birds.

Some migration routes in middle/late Spring (before nesting season) are more less known [17]. As well as geographical abundance of species or nesting site can be found in birds atlas or citizens participatory ornithological projects [17].

4.2. Meat food chains analysis

We propose an analysis of possible contamination of meat/eggs imported from Ukraine or from farms/slaughterhouses in Lubelskie province from May (if founded). Example of supply chains can be easily found in the Internet [17] and are available in ministry of agriculture registry.

4.3. Extended epidemiological analysis

We recommend to perform active monitoring (PCR and serology) of water birds in Pomerania and selected sites in Western Poland (as maybe some low viral pressure of active virus is still present at the end of July 2023). Retrospective investigation of south-eastern border area should be done to verify id Lublin cases as the origin of the outbreak. Possible excess mortality rates from veterinary clinics in May on Polish Ukrainian border should be analyzed.

Regression analysis for risk factors among positive and negative results should be performed. Such a study would report on the spatio-temporal dynamics and the factors that might have influenced A/H5N1 in cats that will be estimated in Poland.

4.4. preparedness for future outbreaks

Measures of control and prevention as well as outbreak investigation in response A/H5N1 in mammals differs significanly between counties in EU (compare Poland [3] vs Finland [29]) as well as intra-country (i.e. Gdańsk metropolitan area vs other regions of Poland). We found that surveillance communication standards for WOAH and WHO/FAO are the most efficient, while ECDC/EFSA (i.e. due to multiple errors in their communicates, not being possible to use electronic disease information system for cats as a host of AI).

We proved, that in case of not-known disease or spillover of known disease to a new host the European Surveillance System (i.e. acting in Poland) is inefficient and needs reorganisation for future threats. This mean that strong national and regional Veterinary and Sanitary authorities are the key resources for new threats [30]. We recommend to shift more responsibilities from ECDC or EFSA to national and sub-national one health centres (with funds and possibility of acting). Thus, national Epidemic Intelligence System aims at identifying, monitoring and analyzing signals of sanitary dangers in human/animal/plant health threatening the Polish territory, in order to produce sanitary information for:

- risk assessment
- risk management
- information on professional channels

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