# EVOLUTION OF THE 2005–2006 AVIAN INFLUENZA H5N1 EPIDEMIC IN ROMANIA: GIS AND SPATIAL ANALYSIS

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## Summary

To provide insights into the spread of H5N1 in Romania during the 2005-2006 epidemic, we used a GIS and spatial analytical methods. Outbreaks first appeared (Nov–Dec) in the eastern part of Romania, particularly within an area that forms part of the Danube River delta. During Feb–Mar, villages in eastern and southern Romania were affected. The largest phase of the epidemic, occurring from May 12 to June 6, affected villages in all parts of central, southern and eastern Romanian. Interpolation facilitated identification of the central role of the environment and landscape (specifically the Danube River delta) in the introduction and initial spread of the epidemic.

## Introduction

Outbreaks of transboundary diseases, such as highly pathogenic avian influenza, provide a unique insight into how animal disease spread through space and time. Spatial epidemiology strives to find interesting patterns in disease occurrence (exploratory spatial data analysis), to show these interesting patterns (data visualization), and to explain these patterns (risk factor analysis and spatial modeling). The application of geographic information systems and spatial statistical methods are critical in revealing subtle patterns of disease spread. Through the information generated, a better understanding of the underlying spatial disease process can be gained, facilitating the design of control and prevention programs and surveillance systems.

H5N1 is an emerging issue for world health: the pathogenic H5N1 influenza strain has caused numerous disease outbreaks in domestic poultry and wildbird populations, and threatens human health. Avian influenza virus infection is endemic in a range of free-living bird species world-wide(1), particularly species associated with water(3). There is a fear that H5N1 could become the next pandemic influenza strain. The aim of this study was to understand how highly pathogenic avian influenza H5N1 might have spread through Romania during the 2005–2006 epidemic.

## Materials and Methods

The first outbreak of H5N1 highly-pathogenic avian influenza was detected in Romania in early October, 2005. The last outbreak was reported in early June, 2006. Data available for confirmed outbreaks of H5N1 included the outbreak location (X, Y coordinates), county and date of occurrence. Reported outbreaks were mapped in *ArcGIS 9.0* (www.esri.com) using a Romanian county shape file (Dealul Piscului 1970 Datum, Stereographic 70 Projection). The mean center and deviational ellipse of these outbreaks was calculated with the relevant functions of the *ArcGIS Spatial Statistics*

*Tools*, these descriptive statistics were also estimated, weighted by the number of days since the first outbreak was reported (7 October 2005). The effect of the weighting scheme used was to emphasize those outbreaks occurring during the latter phases of the epidemic. In addition, descriptive statistics were also estimated for each of the epidemic phases identified.

The spatio-temporal distribution of outbreaks was described by Moran’s autocorrelation statistic in *ArcGIS 9.0 Spatial Statistics Tools*, in which the outcome of interest was outbreak day. Variography was also used to visualize the spatio-temporal distribution of the epidemic. All possible unique pairs of outbreak locations (n= 12,880) were formed in *Variowin 2.0* (www-sst.unil.ch/research/variowin/index.html). A matrix of Euclidean distances and outbreak days was calculated and a semivariogram was formed. A range of lag numbers and lag spacings were chosen to produce a semivariogram which could be described by one of a number of *a priori* models. Several statistical models (exponential, Gaussian, spherical, power) were investigated to identify a line of best fit. Using the line-of-best-fit approach, the parameters of the selected model (nugget, range, sill) were estimated. These parameters were used to produce an interpolated map of outbreak day with *ArcGIS 9.0 Spatial Analyst Extension*. These maps were overlaid on the location of outbreaks to identify localities where outbreaks were reported early during the epidemic. Local clusters of outbreaks were investigated by estimating Anselin’s local indicator of spatial autocorrelation statistic in *ArcGIS 9.0 Spatial Statistics Tools*. Spatial weights were defined by the inverse of Euclidean distance squared, and were globally standardized.

## Results

During the epidemic period (243 days), a total of 165 outbreaks were detected in 25 of the 41 counties (*judete*) of Romania. The largest number of outbreaks was reported from central Romania (*Figure 1*). The median epidemic date was 18 May, day 224 of the epidemic. Half of all outbreaks were reported during a 85-day period, between 27 February and 23 May (epidemic days 144–229). Outbreaks reoccurred at 4 locations.

The mean center of all reported outbreaks in Romania was 29 km northeast of the mean center of outbreaks weighted by outbreak day. Standard deviational ellipses calculated for these centers were essentially the same shape and overlapped. Visual inspection of the epidemic curve revealed 3 epidemic phases: 7 October – 29 December 2005 (days 1–84), 30 December 2005 – 16 April 2006 (days 85–192), and 17 April – 6 June 2006 (days 193–243). The mean centers of outbreaks reported in the first and second epidemic phases were located 43 km apart in eastern Romania (*Figure 1*). The mean center of outbreaks reported in the third epidemic phase was located in central Romania, 58 km northwest of the mean center of all reported outbreaks (*Figure 1*).

The semivariogram of outbreak day, using 15 lags and a lag size of 25 km (Figure 2) was best fit by a spherical model, with an estimated nugget, range and partial sill of 3.6, 190 km and 4.32, respectively. The resulting map of interpolated outbreak day (*Figure 3*) clearly shows the occurrence of H5N1 outbreaks in eastern Romania during the first 120 days of the epidemic.

The distribution of outbreaks was strongly clustered (I = 0.74, P < 0.001) by outbreak day. Clusters of outbreaks in southeastern Romania were identified by the local indicator of spatial autocorrelation (LISA) statistic (*Figure 4*). The principal cluster was located in Braila county, in which 9 outbreaks occurred within a distance of 24 km between 20 November 2005 and 7 January 2006 (epidemic days 45 and 93).

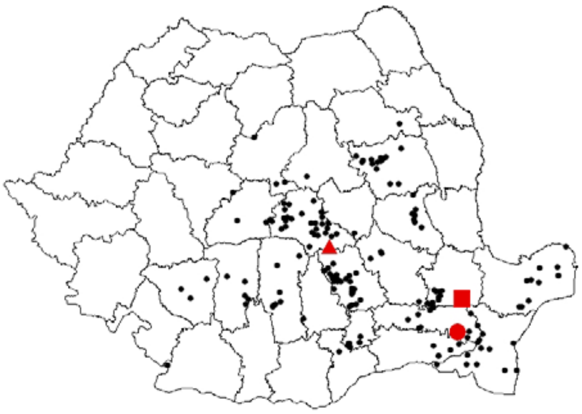

**Figure 1** Distribution of H5N1 highly-pathogenic avian influenza outbreaks (•) in Romania, October 2005 – June 2006. Mean centers of the 3 epidemic phases are shown: days 1–84 (■), days 85–192 (●), and days 193–243 (▲).

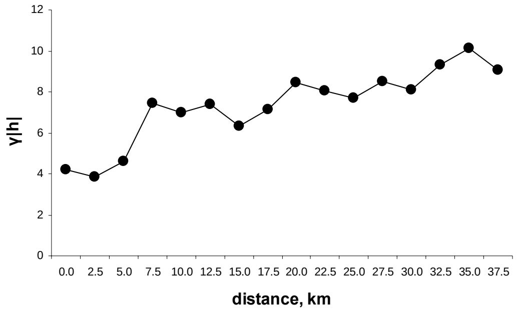

**Figure 2** Semivariogram of the day of H5N1 highly-pathogenic avian influenza outbreaks in Romania, October 2005 – June 2006.

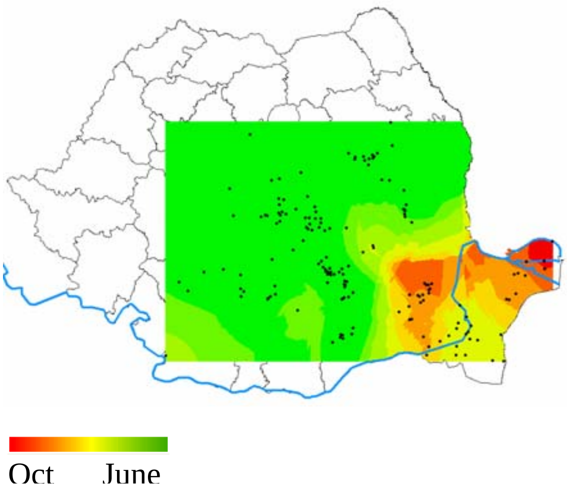

**Figure 3**. Interpolated day of H5N1 highly-pathogenic avian influenza outbreaks in Romania, October 2005 – June 2006. The Danube River is shown.

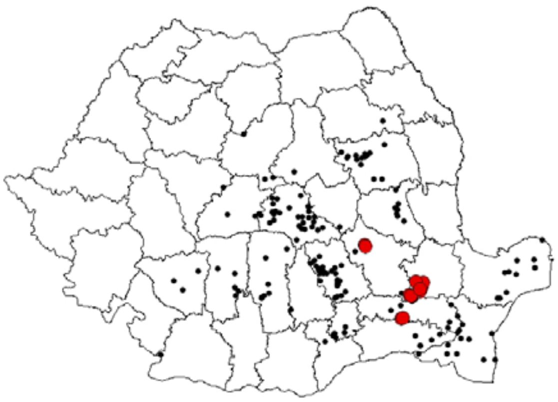

**Figure 4**. Local clusters (●) of H5N1 highly-pathogenic avian influenza outbreaks (•) in Romania, October 2005 – June 2006, identified by the LISA statistic.

## Discussion

Avian influenza outbreaks – both high and low pathogenic – in poultry are often assumed to result from exposure to wild birds. Waterfowl and shorebirds can be infected by all subtypes of influenza A viruses with few or no clinical signs(4). These species are probably responsible for the spread of viruses between regions(2). In the northern hemisphere, influenza virus infection rates are highest during spring migration for shorebirds, whereas waterfowl infections peak in late summer and early fall(4).

The epidemic of H5N1 that occurred in Romania initially developed in the eastern part of the country. This area of Romania is unique: it is bordered to the east by the Black Sea and is characterized by the Danube River delta. There is an extensive wetland system, forming a major breeding area and point of congregation for migratory birds on the Black Sea–Mediterranean flyway (which extends from west Africa to central Asia). The spatio-temporal distribution of outbreaks suggests that H5N1 was introduced by migratory waterfowl during the autumn of 2005. Infection appears to have been mostly confined to this region until May 2005, when a large epidemic of 110 outbreaks occurred mostly in central Romania. If H5N1 was introduced to Romania via migratory waterfowl, winter conditions might have limited its spread. Thus, only localized outbreaks occurred as the result of contact between wild birds and domestic village poultry at common feeding sites, such as ponds and lakes. It is possible that contact between wild birds (both migratory and non-migratory waterfowl and shorebirds) and village poultry, and human factors, might have promoted the large epidemic that occurred during May and June, 2006. Since 6 June, 2006, no outbreaks of H5N1 have been reported in Romania.

An important tool that geographic information systems offer is data visualization. Locations of disease outbreaks may simply be plotted as a point map, or aggregated to form a choropleth map. However, such methods may prevent adequate visualization and hypothesis generation and testing, and lead to inefficient resource allocation for surveillance. Smoothing and interpolation methods reduce the artificial effects of administrative boundaries and facilitate identification of patterns by estimating disease occurrence at a given location using data from surrounding locations. In this study, interpolation facilitated identification of the central role of the environment and landscape (specifically the Danube River delta) in the introduction and initial spread of H5N1 avian influenza in Romania during the 2005–2006 epidemic.

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