Global epidemiology of avian influenza A H5N1 virus infection in humans, 1997–2015: a systematic review of individual case data

Shengjie Lai*, Ying Qin*, Benjamin J Cowling*, Xiang Ren, Nicola A Wardrop, Marius Gilbert, Tim K Tsang, Peng Wu, Luzhao Feng, Hui Jiang, Zhibin Peng, Jiandong Zheng, Qiaohong Liao, Sa Li, Peter W Horby, Jeremy J Farrar, George F Gao, Andrew J Tatem, Hongjie Yu

Lancet Infect Dis 2016

Published **Online**
May 17, 2016
http://dx.doi.org/10.1016/S1473-3099(16)00153-5

*Contributed equally

Avian influenza A H5N1 viruses have caused many, typically severe, human infections since the first human case was reported in 1997. However, no comprehensive epidemiological analysis of global human cases of H5N1 from 1997 to 2015 exists. Moreover, few studies have examined in detail the changing epidemiology of human H5N1 cases in Egypt, especially given the outbreaks since November, 2014, which have the highest number of cases ever reported worldwide in a similar period. Data on individual patients were collated from different sources using a systematic approach to describe the global epidemiology of 907 human H5N1 cases between May, 1997, and April, 2015. The number of affected countries rose between 2003 and 2008, with expansion from east and southeast Asia, then to west Asia and Africa. Most cases (67·2%) occurred from December to March, and the overall case-fatality risk was 483 (53·5%) of 903 cases which varied across geographical regions. Although the incidence in Egypt has increased dramatically since November, 2014, compared with the cases beforehand, there were no significant differences in the fatality risk, history of exposure to poultry, history of patient contact, and time from onset to hospital admission in the recent cases.

Introduction

Highly pathogenic avian influenza A H5N1 virus was first isolated from and characterised in a domestic goose in Guangdong province, China, in 1996,[1] and outbreaks have since been reported in domestic poultry, wild birds, and humans in more than 60 countries.[2–4] The spread of highly pathogenic avian influenza A H5N1 virus in poultry populations increases the risk of human infections.[5–8] The first reported case of human illness caused by H5N1 infection occurred in May, 1997, in Hong Kong, China, with a total of 18 cases and six deaths.[9–12] After an apparent 5-year absence, two cases of H5N1 infection in people who had a history of travel to southern China were reported in February, 2003, in Hong Kong.[13] Following the pattern of spread and persistence of the virus in poultry, human cases of H5N1 infection with high mortality were subsequently detected in China,[14,15] southeast Asia,[16,17] west Asia,[18,19] and Africa, with cases detected in Egypt every year from 2006 to 2015.[20–23] Compared with previous years,[24–26] the incidence of human H5N1 cases has remained at a low level worldwide between October, 2012, and October, 2014,[27,28] and attention has focused on the emergence of variant swine influenza A H3N2 virus in North America,[29] avian influenza A H7N9 virus in China,[30–32] other avian influenza A H5 virus subtypes in Asia, Europe, and North America,[27,33] and other emerging infections.[34–36]

However, between Nov 1, 2014, and April 30, 2015, 165 cases of human H5N1 infection (including 48 deaths) were reported to WHO.[37] This is, by far, the highest number of human cases ever reported globally over a similar period.[38] Moreover, the number of human H5N1 cases reported in Egypt in February, 2015, is the highest number reported by any country in a single month.[39] Emergence of a new cluster of H5N1 clade 2.2.1.2 has been found in poultry in Egypt since mid-2014 and has quickly become predominant.[40] It is not yet known whether or not this emerging phylotype has increased zoonotic potential and, thus, can be transmitted more efficiently to humans.[39–41]

Comprehensive epidemiological analysis of global human cases of H5N1 from 1997–2015 is scarce,[17,42–45] and few studies have presented in detail the changing epidemiology of human H5N1 cases in Egypt by comparing the cases before November, 2014, with the most recent outbreaks from November, 2014, to April, 2015.[20,40,46] To improve understanding of the epidemiology of highly pathogenic avian influenza H5N1 virus, we did a systematic review of individual case data to describe the magnitude and distribution of all human H5N1 cases globally with illness onset between May 1, 1997, and April 30, 2015. We focused on the characteristics of patients, seasonal and geographical patterns, and examined in more detail the epidemiology of human H5N1 cases in Egypt.

Methods

Search strategy and selection criteria

Human H5N1 case data were identified and compiled according to the probable and confirmed case definitions described below. Data on all human H5N1 cases in mainland China were downloaded from the online National Notifiable Infectious Disease Reporting Information System at the Chinese Center for Disease Control and Prevention.[30,47] Data on human H5N1 cases in Vietnam and Azerbaijan as of April 30, 2014, were provided by the Vietnam National Institute of Hygiene and Epidemiology and the Azerbaijan Ministry of Health. Data on human H5N1 cases in all other affected countries or regions were obtained from publicly available sources (appendix), including the WHO Disease Outbreak News of the Global Alert and Response,[48] the WHO Weekly Epidemiological Record,[49] the WHO Western Pacific

Division of Infectious Disease, Key Laboratory of Surveillance and Early-warning on Infectious Disease, Chinese Center for Disease Control and Prevention, Beijing, China (S Lai MD, Y Qin PhD, X Ren MD, L Feng PhD, H Jiang MD, Z Peng MD, J Zheng PhD, Q Liao MD, S Li MD, Prof H Yu PhD); **Department of Geography and Environment, University of Southampton, Southampton, UK** (S Lai, N A Wardrop PhD, Prof A J Tatem PhD); **Flowminder Foundation, Stockholm, Sweden** (S Lai, Prof A J Tatem); **WHO Collaborating Centre for Infectious Disease Epidemiology and Control, School of Public Health, Li Ka Shing Faculty of Medicine, The University of Hong Kong, Hong Kong Special Administrative Region, China** (Prof B J Cowling PhD, T K Tsang MPhil, P Wu PhD); **Biological Control and Spatial Ecology, Université Libre de Bruxelles, Brussels, Belgium** (M Gilbert PhD); **Fonds National de la Recherche Scientifique, Brussels, Belgium** (M Gilbert); **Oxford University Clinical Research Unit, Wellcome Trust Major Overseas Programme, Ho Chi Minh City, Vietnam** (Prof P W Horby MD, Prof J J Farrar MD); **Centre for Tropical Medicine, Nuffield Department of Clinical Medicine, University of Oxford, Oxford, UK** (Prof P W Horby, Prof J J Farrar); **Singapore Infectious Disease Initiative, Singapore** (Prof P W Horby, Prof J J Farrar); **International Severe Acute Respiratory and Emerging Infection Consortium, Centre for Tropical Medicine, University of Oxford,**

Churchill Hospital, Oxford, UK (Prof J J Farrar); **CAS Key Laboratory of Pathogenic Microbiology and Immunology, Institute of Microbiology, Chinese Academy of Sciences, Beijing, China** (Prof G F Gao PhD); **Chinese Center for Disease Control and Prevention, Beijing, China** (Prof G F Gao); **Fogarty International Center, National Institutes of Health, Bethesda, MD, USA** (Prof A J Tatem); **and School of Public Health, Fudan University, Key Laboratory of Public Health Safety, Ministry of Education, Shanghai, China** (Prof H Yu)

Correspondence to:
Prof Hongjie Yu, School of Public Health, Fudan University, Key Laboratory of Public Health Safety, Ministry of Education, Xuhui District, Shanghai 200032, China
cfetpyhj@vip.sina.com

For the **Chinese Center for Disease Control and Prevention** see http://www.cdpc.chinacdc.cn/

For more on **ProMed-mail** see http://www.promedmail.org/

See Online for appendix

Region's Avian Influenza Weekly Update,[50] the FluTrackers website,[51] and the websites of the ministries of health in individual countries or regions.

We also searched PubMed for related studies using a systematic review approach that followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines (figure 1).[52] Articles published from May 1, 1997, to April 30, 2015, were indentified with the query "(H5N1[Title] AND (PATIENT[Title] OR PATIENTS[Title] OR HUMAN[Title] OR HUMANS[Title] OR PERSON[Title] OR CASE[Title] OR CASES[Title])) AND ("1997/05/01"[Date-Publication]: "2015/04/30"[Date-Publication])". Articles published in English and Chinese were included, and full-text Chinese articles were found by searches of China National Knowledge Infrastructure and Wanfang Data. Relevant articles published between 1997 and 2015 were identified through searches in reports from WHO and ProMed-mail posts. Articles resulting from these searches and relevant references cited in those articles were reviewed.

WHO Disease Outbreak News of the Global Alert and Response updates and WHO statistics on the cumulative number of confirmed human H5N1 cases from November, 2003, to April, 2015, were used to establish a list of human H5N1 cases.[48,53] All cases from sources other than WHO updates were matched with the initial list (figure 1). New cases, which were not yet officially announced by WHO, were identified with ProMed-mail posts and FluTrackers, and confirmed by the announcements of ministries of health in individual countries or regions. When crucial information was missing, additional information was sought from published literature, ProMed-mail posts and English language news releases from the regional office of WHO and the relevant ministry of health (appendix p 1).[18,50,54–56]

Case definition

WHO case definition was used to define cases of H5N1 infection.[57] A confirmed case was defined as a human case of influenza A H5N1 virus infection reported by WHO and with laboratory confirmation—ie, a patient with defined clinical signs, epidemiological linkage, and laboratory confirmation by an influenza laboratory accepted by WHO, as specified in WHO case definition. Other reported cases were considered as probable cases if they had exposure to other confirmed patients or to sick or dead poultry, or the H5N1 infection was confirmed by the country or local institutions, but did not meet WHO criteria or was not announced by WHO.

Data variables and extraction

All probable and confirmed cases with illness onset by April 30, 2015, were included in the analysis. Individual data on cases included age, sex, country, type of diagnosis, year, month and day of onset, date of hospital admission, final outcome (fatal or non-fatal), date of outcome, and potential risk factors (appendix pp 8–9). Information on exposure potentially related to the acquisition of H5N1 infections was collected (panel). Where contradictory information was found for a given variable, WHO and ministry of health data were given priority over journal articles, and journal articles were given priority over other

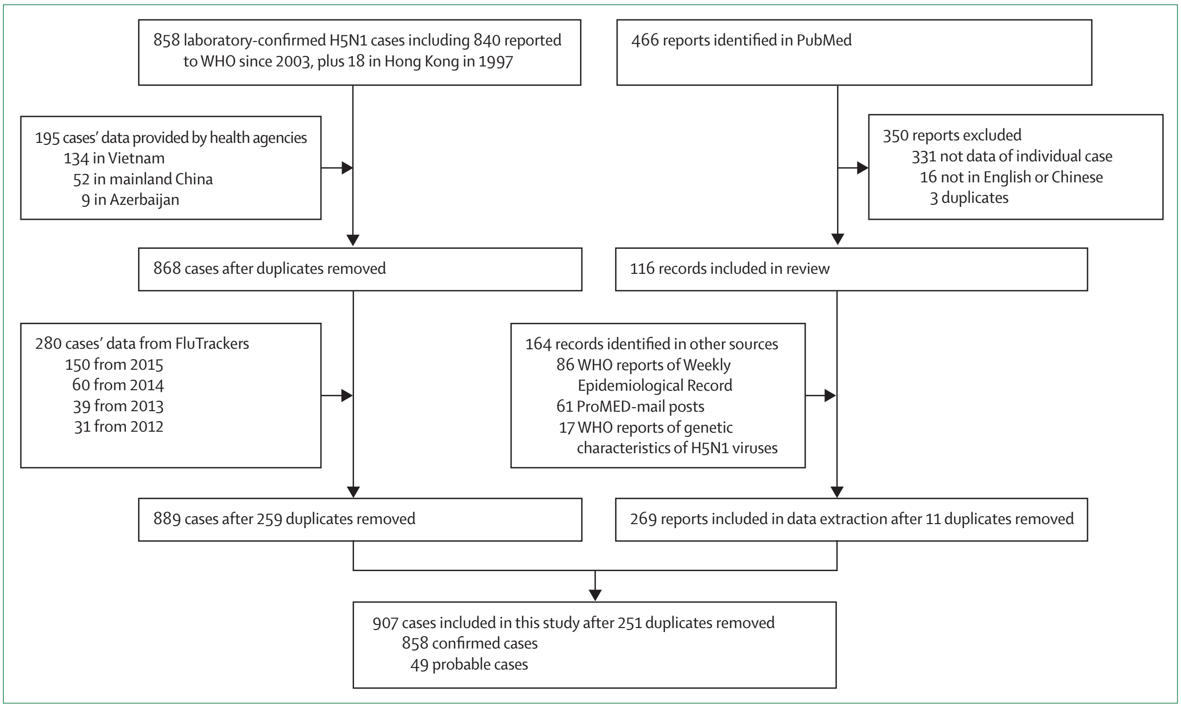

Figure 1: **Study selection and collection of individual case data**

sources of information (appendix p 1). Epidemic curves were plotted and the patient characteristics were summarised by outcome and geographical region.

Data on the clade or subclade of H5N1 isolated from patients were collated from the regular WHO reports.[58] 17 reports issued between August, 2006, and February, 2015, were reviewed, which provided information on H5N1 clades circulating and characterised from 1997 to February, 2015.[58] However, not all individual cases were reported with laboratory results of the clade or subclade. Where this information was not available, the infection was presumed to be in the clade or subclade of H5N1 that was present in the same period and area.[17,40,42,59,60] All data used in this study were anonymised.

Ethical approval

The National Health and Family Planning Commission of China, the Ministry of Health of Vietnam, and the Ministry of Health of Azerbaijan determined that the collection of data from human cases of avian influenza A H5N1 virus infection was part of the public health investigation of an outbreak and was exempt from institutional review board assessment. All other data from other countries were obtained from publicly available data sources. All data were supplied and analysed in an anonymous format, without access to personal identifying information.

Results

907 human cases of H5N1 infection were reported globally between May 1, 1997, and April 30, 2015, of which 94·6% were confirmed cases and 5·4% were probable cases (table 1, figure 2). The number of cases per year varied, with the highest numbers recorded in 2015 (figure 3, appendix p 18, 19). The total number of cases (n=213) in 2014–15 was greater than that recorded from 2010–13 (n=181; appendix p 19), with the highest number of cases recorded in February, 2015, when 55 cases were reported in Egypt and one in China.

The overall male-to-female ratio was almost even (1:1·2) from 1997 to 2014, although this pattern was not uniform across regions (table 1). The median age of patients was 19 years (IQR 5–32), and 363 (41·2%) of 881 cases were in children younger than 15 years and 707 (80·3%) of 881 cases were in people younger than 35 years. The median age of patients who had non-fatal cases of H5N1 infection was lower in north Africa and older in east and southeast Asia (median 6 years [IQR 3–31] *vs* 18·5 years [6–30]), but the median age of patients who had fatal cases was higher in north Africa than in east and southeast Asia (30 years [20–36] *vs* 19 years [9–30]; figure 4).

16 countries reported human cases between 1997 and 2015. The number of affected countries has risen between 2003 and 2008, with expansion from east Asia to southeast Asia, then west Asia, north Africa, and other regions, and apparent ongoing transmission and cases reported almost every year in China, Vietnam, Cambodia, Indonesia, and Egypt (appendix p 20). The incidence in Asia remained at low levels in 2013–15, while the number of cases in Egypt has increased in 2014–15. During 1997–2015, 594 (67·2%) of 884 cases were reported between December and March, with a peak in January (20·9%; appendix p 21). However, compared with countries in southeast Asia and north Africa, countries in east Asia and west Asia had fewer cases in the warm or hot season from April to September (8·1% *vs* 26·2%), and showed earlier peaks (December *vs* January) and shorter epidemic periods, with cases occurring year-round in southeast Asia and north Africa, but from January to June and October to December in east Asia, and only from December to March in west Asia (appendix p 20, 21).

Panel: **Definition of exposures to poultry and humans**

Occupational exposure to live poultry
Poultry-related exposure at place of work (eg, people involved in the breeding, trafficking, sale, and quarantine of poultry) in the 2 weeks before symptom onset

Exposure to poultry at live bird markets
Visit of a wholesale or retail market of live poultry or birds in the 2 weeks before symptom onset

Exposure to sick or dead poultry
Direct physical contact with, or proximity to, sick or dead poultry or poultry products (eg, meat) or faeces in the 2 weeks before symptom onset

Exposure to backyard poultry
Direct physical contact with, or proximity to poultry raised in the backyard within 2 weeks before symptom onset

Any exposure to poultry
Direct or indirect contact with, or proximity to healthy, sick, or dead poultry (including any kind of poultry or birds—eg, chickens, ducks, geese, pet birds, pigeons) in live bird markets, backyards, farms, or neighbourhoods, or consumption of improperly processed poultry products

Exposure to virus through patient contact
A patient with a history of close contact with a person with confirmed or probable influenza H5N1 virus infection (at any time from the day before symptom onset to death, or during the period that the patient was admitted into hospital) in the 2 weeks before symptom onset

	Total (n=907)	East and southeast Asia (n=505)	North Africa (n=363)	Other (n=39)
Type of case				
Confirmed	858 (94·6%)	479 (94·9%)	343 (94·5%)	36 (92·3%)
Probable	49 (5·4%)	26 (5·1%)	20 (5·5%)	3 (7·7%)
Sex				
Female	476 (52·5%)	246 (48·7%)	213 (58·7%)	17 (43·6%)
Unknown	29 (3·2%)	21 (4·2%)	6 (1·7%)	2 (5·1%)
Age, years				
Median (IQR)	19 (5–32)	19 (8–30)	20 (4–34)	15 (5–22)

(Table 1 continues on next page)

	Total (n=907)	East and southeast Asia (n=505)	North Africa (n=363)	Other (n=39)
(Continued from previous page)				
Final outcome				
Death	483 (53·3%)	349 (69·1%)	116 (32·0%)	18 (46·2%)
Unknown	4 (0·4%)	2 (0·4%)	2 (0·6%)	0
Hospital admission				
Yes	819 (90·3%)	438 (86·7%)	353 (97·2%)	28 (71·8%)
Unknown	82 (9·0%)	64 (12·7%)	9 (2·5%)	9 (23·1%)
Time delay from onset to hospital admission, days				
Median time (IQR)	4 (2–6)	5 (3–7)	3 (1–6)	2 (1–5)
Unknown	184 (20·3%)	121 (24·0%)	46 (12·7%)	17 (43·6%)
Time delay from hospital admission to death or discharge (recovery), days				
Median time (IQR)	5 (2–9)	4 (2–9)	5 (2–9)	5 (3–18)
Unknown	403 (44·4%)	166 (32·9%)	219 (60·3%)	18 (46·2%)
Time delay from onset to death or discharge (recovery), days				
Median time (IQR)	10 (7–15)	10 (7–15)	10 (7–14)	9 (7–20)
Unknown	360 (39·7%)	124 (24·6%)	221 (60·9%)	15 (38·5%)
Predominant clade or subclade				
0	18 (2·0%)	18 (3·6%)	0	0
1	193 (21·3%)	193 (38·2%)	0	0
2.1	208 (22·9%)	208 (41·2%)	0	0
2.2	393 (43·3%)	0	363 (100%)	30 (76·9%)
2.3	89 (9·8%)	84 (16·6%)	0	5 (12·8%)
7	2 (0·2%)	2 (0·4%)	0	0
Unknown	4 (0·4%)	0	0	4 (10·3%)
Exposure history				
Any exposure to poultry				
Exposure	748 (82·5%)	382 (75·6%)	339 (93·4%)	27 (69·2%)
Unknown	126 (13·9%)	94 (18·6%)	24 (6·6%)	8 (20·5%)
Occupational exposure to live poultry				
Exposure	15 (1·7%)	12 (2·4%)	2 (0·6%)	1 (2·6%)
Unknown	586 (64·6%)	289 (57·2%)	286 (78·8%)	11 (28·2%)
Visits to live bird markets				
Visits	82 (9·0%)	68 (13·5%)	11 (3·0%)	3 (7·7%)
Unknown	596 (65·7%)	296 (58·6%)	286 (78·8%)	14 (35·9%)
Exposure to sick or dead poultry				
Exposure	439 (48·4%)	242 (47·9%)	174 (47·9%)	23 (59·0%)
Unknown	395 (43·6%)	217 (43·0%)	166 (45·7%)	12 (30·8%)
Exposure to backyard poultry				
Exposure	188 (20·7%)	113 (22·4%)	64 (17·6%)	11 (28·2%)
Unknown	601 (66·3%)	301 (59·6%)	286 (78·8%)	14 (35·9%)
Human case contact				
Contact	49 (5·4%)	35 (6·9%)	3 (0·8%)	11 (28·2%)
Unknown	115 (12·7%)	86 (17·0%)	21 (5·8%)	8 (20·5%)

Data are presented as n (%) of patients unless otherwise indicated. East and southeast Asia (505 cases): Indonesia (208), Vietnam (134), Cambodia (58), mainland China (52), Thailand (27), Hong Kong (23), Laos (2), and Myanmar (1). North Africa (363): Egypt (363). Other (39): Turkey (12), Azerbaijan (9), Bangladesh (7), Pakistan (4), Iraq (3), Nigeria (2), Djibouti (1), and Canada (1). Data on H5N1 clade or subclade were based on the reports from WHO website or scientific literature, and the known geographic distribution of the viruses. Not all cases were laboratory confirmed and reported with clade results, so we presumed that the case in each area was infected by the reported predominant clade or subclade of H5N1 in the same period and area. The clade or subclade in each area were clade 0 in Hong Kong in 1997; clade 1 in Vietnam, Cambodia, Thailand, and Hong Kong; subclade 2.1 mainly in Indonesia; subclade 2.2 in Egypt, Turkey, Azerbaijan, Bangladesh, Iraq, Nigeria, and Djibouti; subclade 2.3 in Vietnam, Bangladesh, Laos, Canada, and Myanmar; and clade 7 in mainland China. Clade data were unavailable for four cases in Pakistan in 2007.

***Table 1:* Characteristics of patients with H5N1 infection by geographic region, May, 1997, to April, 2015**

After exclusion of four cases with unknown outcome (two from Vietnam in 2005 and two from Egypt in 2015), the overall case-fatality risk was 53·5% (483 of 903 cases), with a decrease from 70·7% (275 of 420 cases) in 2003–08 to 43·4% (202 of 465 cases) in 2009–15, and varied across geographical regions, with a case-fatality risk (69·4%, 349 of 503 cases) in east and southeast Asia more than two times that in north Africa (32·1%, 116 of 361 cases; table 1). The age distribution of patients also differed by outcome, with a median age of 22 years (IQR 12–32) for patients with fatal cases and 10 years (3–30) in patients who recovered (figure 3). Most cases (95·8%, 748 of 781) reported exposure to poultry including 85·7% (439 of 512 patients) exposed to sick or dead poultry, 61·4% (188 of 306 patients) exposed to backyard poultry, 26·4% (82 of 311 patients) exposed to poultry at live bird markets, and 4·7% (15 of 321 patients) occupationally exposed to live poultry. Additionally, 6·2% (49 of 792 patients) reported contact with a patient with human H5N1 infection before the onset of illness (table 1, appendix p 10). Time from onset of illness to hospital admission was available for 79·7% (723 of 907 patients) with a median of 4 days (IQR 2–6). Generally, patients who survived were admitted into hospital earlier than patients who died (median 3 days [IQR 1–6] *vs* 5 days [3–7]; appendix). Additionally, patients in north Africa had a shorter time from onset to hospital admission than patients in east and southeast Asia (3 days [1–6] *vs* 5 days [3–7]), but the median time from onset to outcome was the same (10 days) between patients in north Africa and cases in east and southeast Asia.

The influenza A H5N1 viruses in human cases have been characterised as clade or subclade 0, 1, 2.1, 2.1, 2.3, and 7 (table 1, 2; appendix p 10). Clade 1 was first reported in Hong Kong in 2003, and then reported in southeast Asia each year from 2003 to 2014, but subclade 2.1 was only reported in Indonesia since 2005, and subclade 2.2 has circulated in Egypt since 2006 with sporadic reporting in Africa and west Asia. Additionally, subclade 2.3 has been reported in east and southeast Asia since 2005.

From March, 2006, to April, 2015, a total of 363 human cases with influenza A H5N1 virus infection were reported in Egypt with 116 deaths (32%; appendix p 23), of which 51% of cases were reported during the 6 months between November, 2014, and April, 2015 (appendix p 15). The male-to-female ratio was not significantly different between cases before November, 2014, and cases in the period from November, 2014, to April, 2015, but of the latter cases, patients had a higher median age (26 years, IQR 4–38) than before November, 2014 (16 years, 3–30), which was also different for both non-fatal and fatal cases (figure 4). However, the case-fatality risk was not significantly different at 36% (64 of 178 cases) before November, 2014, compared with 28·4% (52 of 183 cases) during November, 2014, to April, 2015

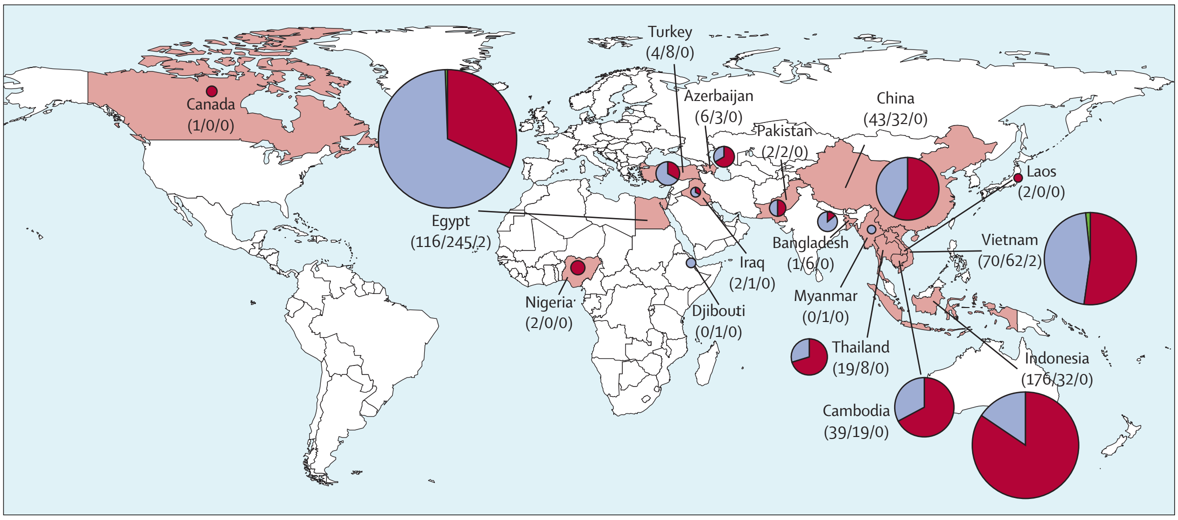

***Figure 2:* Geographic distribution of human cases of H5N1 infection by outcome, May, 1997, to April, 2015 (n=907)**
Data for China include cases reported by mainland China (52 cases) and Hong Kong, China (23 cases). In the pie charts, the blue sectors show the proportion of non-fatal cases, the red fatal cases, and the green unresolved cases. The numbers beneath the country names show numbers of cases: fatal/non-fatal/unresolved.

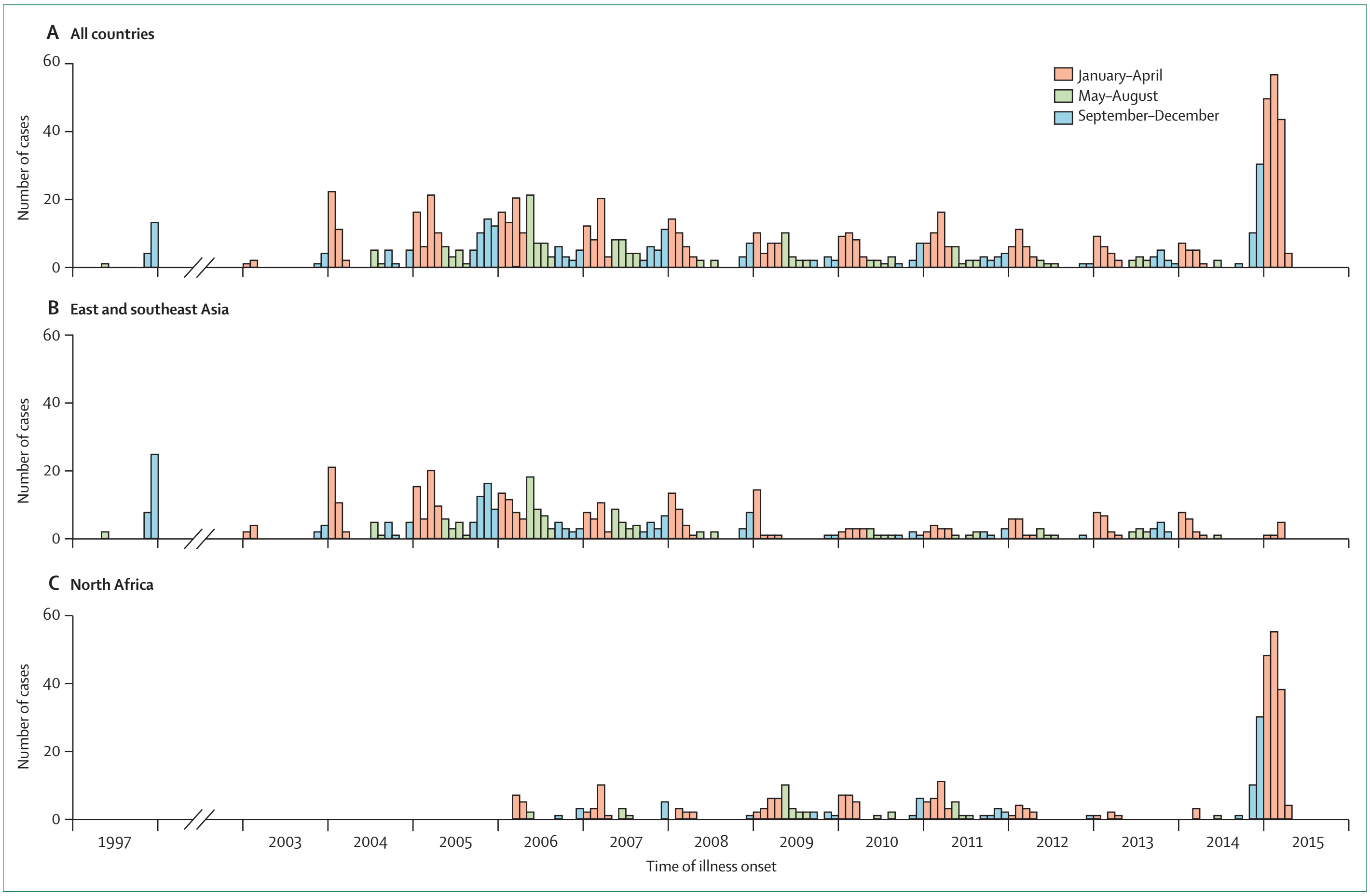

***Figure 3:* Epidemic curve of human cases of H5N1 infection by region, May, 1997, to April, 2015**
(A) H5N1 human cases reported worldwide (884 cases). (B) East and southeast Asian countries (484 cases) were Indonesia (187), Vietnam (134), Cambodia (58), mainland China (52), Thailand (27), Hong Kong (23), Laos (2), and Myanmar (1). (C) North African countries (363 cases) were Egypt (363). Month of illness was unknown in 23 cases (21 cases in Indonesia in 2009 and two cases in Turkey in 2006) and were excluded from this epidemic curve.

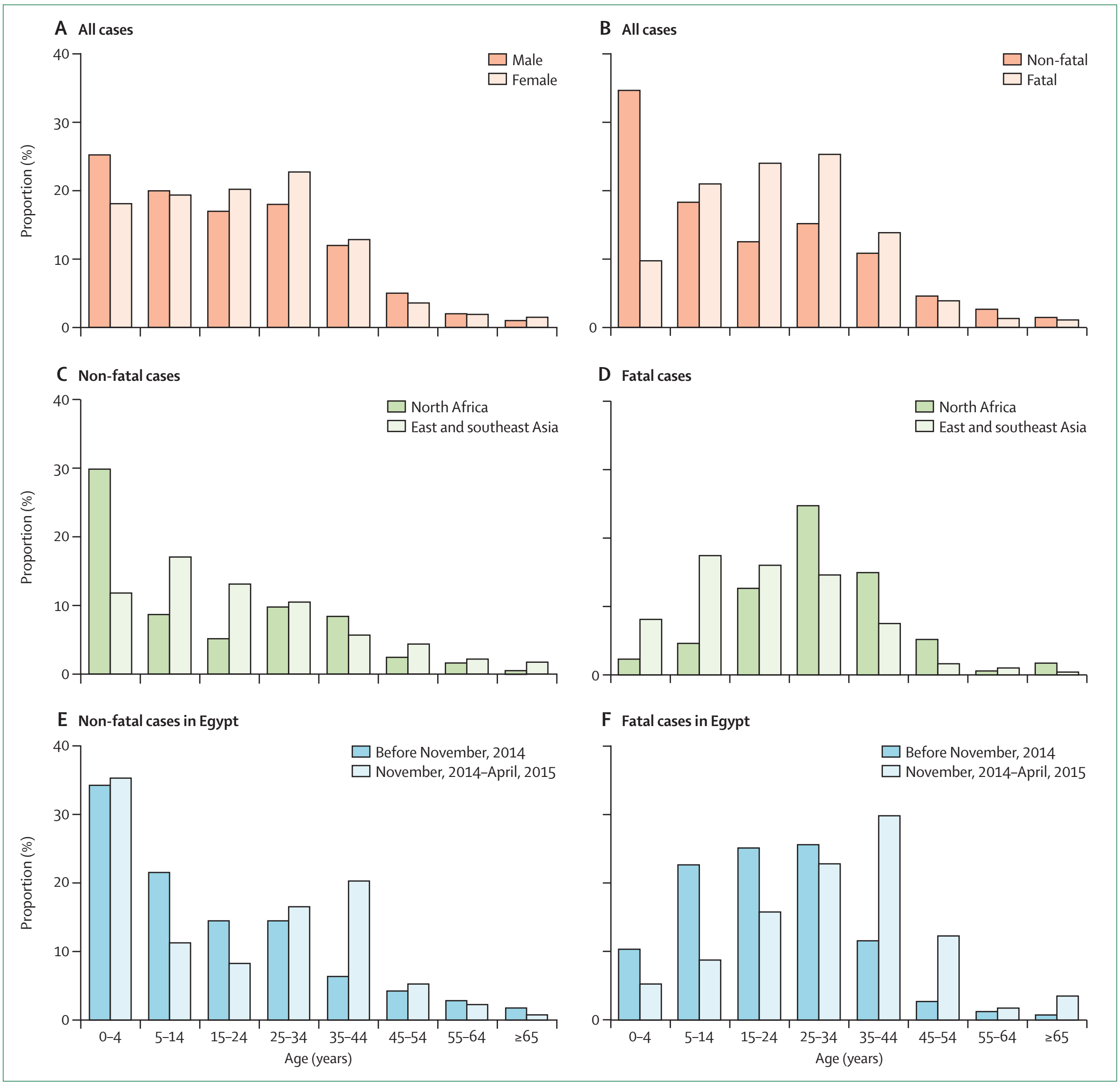

***Figure 4:* Age distribution of human cases with H5N1 infection by gender, geographic regions, and outcome, May, 1997, to April, 2015**
(A) All male (n=401) and female cases (n=476). (B) All fatal (n=463) and non-fatal cases (n=416). (C) Non-fatal cases in north Africa (n=245) and east and southeast Asia (n=152). (D) Fatal cases in north Africa (n=116) and east and southeast Asia (n=329). (E) Non-fatal cases in Egypt before (n=114) and since Nov 1, 2014 (n=131). (F) Fatal cases in Egypt before (n=64) and since Nov 1, 2014 (n=52).

(appendix p 15). For fatal cases, the median time and IQR (5 days, 3–6) for onset to hospital admission was the same from March, 2006, to October, 2014, and November, 2014, to April, 2015, but the time was different for non-fatal cases before November, 2014 (1 day, 1–3), and in November, 2014, to April, 2015, (4 days, 2–6). Most cases reported a history of poultry exposure—96·1% before November, 2014, and 90·8% in November, 2014, to April, 2015.

Discussion

In this Review, a global dataset spanning 18 years was systematically collated to investigate changes in the epidemiological characteristics of human H5N1 cases, and we focused on Egypt, given its unique situation of increasing incidence since November, 2014.[20,37,46] Our findings suggest that the geographical extent of human H5N1 cases has expanded from east Asia to southeast Asia, then to west Asia and north Africa in 2003–09, which could be related to global spread of the virus via bird migration.[61–63] The bird migration network was shown to better reflect the observed viral gene sequence data than other networks (eg, poultry trade networks) and contributes to seasonal H5N1 epidemics in local regions.[3,5,7] Additionally, previous evidence showed Siberia as a major hub for the spread of the virus via birds, and southeast Asia and Africa as areas of local virus persistence and the major sources of genetically and antigenically novel strains.[5,7,64,65] Therefore, the increasing range of virus spread and outbreaks among birds might also increase the risk of human exposure.[3,66] However, some of the apparent geographical spread in cases could also be attributable to enhanced clinical and laboratory surveillance capacity in the past 15–20 years.

Human H5N1 infections were found to exhibit seasonality, related to the cooler season from December to March and across diverse climate zones in the northern hemisphere (appendix p 20, 21), which might correlate with the migration patterns of wild birds and the activity of the virus in winter or cooler seasons.[3,7,43] Investigators of a 2015 study found that the timing of H5N1 outbreaks and viral migrations were closely associated with bird migration networks in Asia.[5] Additionally, the lower temperatures in Asia and north Africa across diverse climates were associated with increasing cases of human H5N1 infection in winter, which is consistent with increased poultry outbreaks and H5N1 transmission during cold and dry conditions, and also overlapped with human seasonal influenza epidemics.[3,43,67,68]

Although most human populations are thought to have little or no immunity to influenza A H5N1 viruses, most patients examined in this study were children and younger adults, and these age groups were also more likely to recover, whereas the fatality risk was higher in adults, which might be related to the immunological reaction of the virus in different age groups.[41] Consistent with previous reports,[28,45] the cases in which time from onset of illness to hospital admission was 3 days or more were more likely to be fatal than those admitted to hospital within 3 days of onset with an odds ratios (OR) of 3·6 (95% CI 2·5–5·1), which might be because of the early administration of antiviral treatment, or selection bias where the patients admitted to hospital later after onset were more likely to be severe.[17] Compared with Indonesia, Vietnam, Cambodia, mainland China, and Thailand, the lower case-fatality risk in Egypt might be related to a less virulent virus clade, less severe clinical disease, and earlier identification, earlier hospital admission, and early treatment with oseltamivir for H5N1 cases.[20,22,44] However, the case-fatality risk might be underestimated because various government entities or reports might not have identified or updated data on which cases had died at the time we collated data. Additionally, almost all human cases of H5N1 infection had close contact (in the 2 weeks before symptom onset) with infected live or dead birds, other human cases, or H5N1-contaminated environments, which reaffirmed reports that human influenza H5N1 virus infection is typically preceded by exposure to sick or dead poultry in backyards, live bird markets, or farms.[69–73]

An increased number of animal-to-human infections has been reported by Egypt during November, 2014, to April, 2015, with the number of cases more than the total of the past 8 years (2006–14).[20] The increase in the number of human cases in Egypt since November, 2014, can be attributed to a mixture of factors, including increased circulation of influenza H5N1 viruses in poultry, lower public health awareness of risks in central and northern Egypt and seasonal factors, such as closer proximity to poultry because of cold weather and possible longer survival of the viruses in the environment.[74] However, the increased numbers of human cases in Egypt are of major concern because of the continued potential pandemic threat from H5N1. A few cases of human-to-human transmission and family clusters have been reported in Egypt and other countries.[40,46,75–79] Nevertheless, influenza H5N1 viruses do not appear to transmit easily from person to person, and the risk of community-level spread of these viruses remains low.[20,27,39]

	First identified, year	Locations identified	Case-fatality risk
Clade 0	1997	Hong Kong	33% (6/18)
Clade 1	2003	Hong Kong, Vietnam, Cambodia, and Thailand	59% (112/191)
Subclade 2.1	2005	Indonesia	85% (176/208)
Subclade 2.2	2005	Turkey, Egypt, Azerbaijan, Djibouti, Iraq, Nigeria, and Bangladesh	33% (130/391)
Subclade 2.3	2005	Mainland China, Laos, Myanmar, Vietnam, Hong Kong, Bangladesh, and Canada	62% (55/89)
Clade 7	2003	Mainland China	100% (2/2)

Data on H5N1 clade or subclade of human cases were based on the reports from WHO website or scientific literature, and the known geographic distribution of the viruses. Not all cases were laboratory confirmed and reported with clade results, so we presumed that the patient was infected by the reported clade or subclade of H5N1 in the same period and area. Clade data were unavailable for four cases in Pakistan in 2007, and four cases with unknown outcome (two in Vietnam in 2005 and two in Egypt in 2015) were also excluded.

***Table 2:* Clade or subclade and fatality of human case with H5N1 infection, May, 1997, to April, 2015**

Influenza H5N1 viruses have evolved from the 1996 progenitor strain and now comprise at least ten clades, through a complexity of genetic changes, which have infected domestic poultry and wild birds in many countries.[1,21,61,62,80] In this study, four clades (0, 1, 2, and 7) and three subclades (2.1, 2.2, and 2.3) of H5N1 strains have infected humans, all of which have been reported in human cases before 2006.[41,61] Compared with clade 0, the cases with clade 1, and subclades 2.1 and 2.3 were more likely to result in death with a crude OR of 2·8 (95% CI 0·93–9·6), 11·0 (3·5–37·8), and 3·2 (1·0–11·4), respectively (appendix p 10). However, the risk of death between cases with clade 0 and subclade 2.1 was not significantly different (OR 1·0, 95% CI 0·3–3·3). Based on the available information, the virus clades isolated from human beings were the same as the clades circulating in local poultry.[21,28] From late 2003 to mid-2005, most H5N1 infections in humans were caused by clade 1 strains in southeast Asia (ie, Vietnam, Thailand, and Cambodia).[61]

Although the highly pathogenic H5N1 strains can be expected to continue evolving over time, preliminary laboratory investigation has not detected major genetic changes in the viruses isolated from the patients or animals in 2014–15 compared with previously circulating isolates in the same regions,[41,81] and the genetic diversity of the influenza H5N1 virus decreased substantially between 1996 and 2011 in China, presumably under strong selective pressure associated with vaccination in poultry.[55] However, other influenza A H5 virus subtypes, such as H5N2, H5N3, H5N6, and H5N8, have been detected in birds in Europe, North America, and Asia, and no human cases of infection have been reported so far, with the exception of three human cases of influenza A H5N6 virus infection detected in China in 2014–15.[39,74] However, the co-circulation of influenza A viruses in human reservoirs and animal reservoirs can provide opportunities for these viruses to reassort and acquire genetic characteristics that facilitate sustained human-to-human transmission—a necessary trait of pandemic viruses.[3,82]

Vaccines and antivirals are the most effective approaches to prevent influenza virus infection and treat illness, respectively.[41,83,84] Vaccination of poultry has been implemented in many of the affected countries to control H5N1 in poultry, especially in those locations where influenza H5N1 viruses have become enzootic in poultry and wild birds.[85–87] 27 H5N1 candidate vaccine viruses for human beings are available and a new candidate vaccine is in preparation to protect against the circulating H5 clade 2.2.1.2 of viruses in Egypt.[41,88] The first adjuvant vaccine for the prevention of influenza H5N1 virus was approved by the US Food and Drug Administration in November, 2013, and this vaccine is being stockpiled for pandemic preparedness by the US Government.[89] Additionally, the antiviral drug oseltamivir can reduce the severity of illness and mortality when started soon after symptom onset and appears to benefit all age groups. Prompt diagnosis and early therapeutic intervention should therefore be considered for all H5N1 cases,[84,90,91] though antiviral resistance continues to receive attention and continued monitoring is needed.[92] The availability of antivirals and vaccines in the event of an H5N1 pandemic should be considered in advance.[93]

There are some limitations to this study. First, the data used were collated from different sources. The data quality might be influenced by key steps in public health surveillance or reports including case definitions, reporting methods, availability of health care and laboratory diagnostics, under-reporting, and the completeness and accuracy of data reported or announced by different countries or organisations. Compared with the areas in which many cases were seen in this study, some countries with few or no cases reported might be attributed to the low availability and capability of public health services, serological testing, and surveillance. Second, detailed data on case characteristics and clinical management were unavailable to assess the association between clinical manifestation, treatment, and outcome, and this study did not include the cases with subclinical H5N1 infection, which have been occasionally reported.[70,94–96] Third, the findings might be affected by missing data on exposure, outcome, and hospital admission, and the misclassification of cases with presumed clade or subclade. Additionlly, this study only included data sources in English or Chinese, which might neglect data on cases reported in other languages, including announcements or reports from Egypt.

In this Review, the high-risk areas, population groups, and seasonality of human highly pathogenic avian influenza H5N1 virus infections have been systematically reviewed, providing evidence for the planning of prevention and control. The geographical distribution of countries with human H5N1 infections has expanded, especially between 2003 and 2008, with variations in outcome, demography, seasonality, and the clade or subclade of viruses across the region. The incidence of human infections increased dramatically in Egypt from November, 2014, to April, 2015, but remained at a low level in other regions, and the case-fatality risk in Egypt has not significantly changed. However, since avian influenza A H5N1 viruses present a continuous threat to human populations, echoing the recommendations of WHO and other organisations on influenza at the human–animal interface,[41,84,97–99] sustained efforts and close collaboration between the animal health sectors and public health sectors at community, national, and international levels to monitor the dynamics in human beings, poultry, and wild birds, and to conduct early clinical management is needed. Downstream research should focus on the development of vaccines and antivirals, explore the driving factors behind the epidemic, and assess the potential for future pandemics.

Contributors

HY designed and supervised the study. SLa and YQ designed the study, collected data, finalised the analysis, wrote the manuscript, and interpreted the findings. XR did analysis and mapped the case geographic distribution. SLi, TKT, LF, HJ, ZP, JZ, and QL did the literature search, data collection, and data analysis. BJC, NAW, MG, PW, PWH, JJF, GFG, and AJT interpreted the findings and commented on and revised drafts of the manuscript. All authors read and approved the final manuscript.

Declaration of interests

BJC has received research funding from MedImmune and Sanofi Pasteur, and is a consultant for Crucell NV. SLa, YQ, BJC, XR, NAW, MG, TKT, PW, LF, HJ, ZP, JZ, QL, SLi, PWH, JJF, GFG, AJT, and HY declare no competing interests.

Acknowledgments

We thank staff members at county-level, district-level, prefecture-level, and provincial-level Centers for Disease Control and Prevention at the provinces with human H5N1 cases for providing assistance with field investigation, administration, and data collection in China; and staff members from WHO, the Ministry of Health of Azerbaijan, and the Vietnam National Institute of Hygiene and Epidemiology for assistance in coordination of data collection. This study was supported by grants from the National Science Fund for Distinguished Young Scholars (81525023); the US National Institutes of Health (Comprehensive International Program for Research on AIDS grant U19 AI51915); the Ministry of Science and Technology of China (2012 ZX10004–201, 2014BAI13B05); the Ministry of Health of China (201202006); China CDCs Key Laboratory of Surveillance and Early-warning on Infectious Disease; the Harvard Center for Communicable Disease Dynamics from the National Institute of General Medical Sciences (U54 GM088558); a commissioned grant from the Health and Medical Research Fund of the Health, Welfare and Food Bureau of the Hong Kong Government; and the University Grants Committee of Hong Kong, China (project number T11–705/14N). SLa is supported by the Flowminder Foundation and the University of Hong Kong for his postgraduate research on population movement and infectious disease dynamics at the University of Southampton. NAW is supported by the UK Medical Research Council (MR/J012343/1). PWH is funded by the Wellcome Trust (grants 089276/Z/09/Z and 089276/B/09/Z) and the EU FP7 project PREPARE (602525). AJT is supported by funding from National Institutes of Health National Institute of Allergy and Infectious Diseases (U19AI089674); the Bill & Melinda Gates Foundation (OPP1106427, 1032350); the RAPIDD program of the Science and Technology Directorate, Department of Homeland Security; and the Fogarty International Center, National Institutes of Health. The funder of the study had no role in the study design, data collection, data analysis, data interpretation, writing of the report, or the decision to publish. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit for publication. The views expressed are those of the authors and do not necessarily represent the policy of the China CDC or the institutions with which the authors are affiliated.

References

1 Duan L, Bahl J, Smith GJ, et al. The development and genetic diversity of H5N1 influenza virus in China, 1996–2006. *Virology* 2008; **380:** 243–54.

2 Olsen B, Munster VJ, Wallensten A, Waldenström J, Osterhaus AD, Fouchier RA. Global patterns of influenza A virus in wild birds. *Science* 2006; **312:** 384–88.

3 Durand LO, Glew P, Gross D, et al. Timing of influenza A(H5N1) in poultry and humans and seasonal influenza activity worldwide, 2004–2013. *Emerg Infect Dis* 2015; **21:** 202–08.

4 World Organisation for Animal Health. Update on highly pathogenic avian influenza in animals (Type H5 and H7). http://www.oie.int/animal-health-in-the-world/update-on-avian-influenza/2015 (accessed Oct 7, 2015).

5 Tian H, Zhou S, Dong L, et al. Avian influenza H5N1 viral and bird migration networks in Asia. *Proc Natl Acad Sci USA* 2015; **112:** 172–77.

6 Chen H, Smith GJ, Zhang SY, et al. Avian flu: H5N1 virus outbreak in migratory waterfowl. *Nature* 2005; **436:** 191–92.

7 Li X, Zhang Z, Yu A, et al. Global and local persistence of influenza A(H5N1) virus. *Emerg Infect Dis* 2014; **20:** 1287–95.

8 Xu X, Subbarao K, Cox NJ, Guo Y. Genetic characterization of the pathogenic influenza A/Goose/Guangdong/1/96 (H5N1) virus: similarity of its hemagglutinin gene to those of H5N1 viruses from the 1997 outbreaks in Hong Kong. *Virology* 1999; **261:** 15–19.

9 Shortridge KF. Poultry and the influenza H5N1 outbreak in Hong Kong, 1997: abridged chronology and virus isolation. *Vaccine* 1999; **17** (suppl 1): S26–29.

10 Subbarao K, Klimov A, Katz J, et al. Characterization of an avian influenza A (H5N1) virus isolated from a child with a fatal respiratory illness. *Science* 1998; **279:** 393–96.

11 Chan PK. Outbreak of avian influenza A(H5N1) virus infection in Hong Kong in 1997. *Clin Infect Dis* 2002; **34** (suppl 2): S58–64.

12 Claas EC, Osterhaus AD, van Beek R, et al. Human influenza A H5N1 virus related to a highly pathogenic avian influenza virus. *Lancet* 1998; **351:** 472–77.

13 Peiris JS, Yu WC, Leung CW, et al. Re-emergence of fatal human influenza A subtype H5N1 disease. *Lancet* 2004; **363:** 617–19.

14 Yu H, Shu Y, Hu S, et al. The first confirmed human case of avian influenza A (H5N1) in Mainland China. *Lancet* 2006; **367:** 84.

15 Normile D. Avian influenza. Panel confirms report of early H5N1 human case in China. *Science* 2006; **313:** 899.

16 Beigel JH, Farrar J, Han AM, et al. Avian influenza A (H5N1) infection in humans. *N Engl J Med* 2005; **353:** 1374–85.

17 Abdel-Ghafar AN, Chotpitayasunondh T, Gao Z, et al. Update on avian influenza A (H5N1) virus infection in humans. *N Engl J Med* 2008; **358:** 261–73.

18 Oner AF, Bay A, Arslan S, et al. Avian influenza A (H5N1) infection in eastern Turkey in 2006. *N Engl J Med* 2006; **355:** 2179–85.

19 Gilsdorf A, Boxall N, Gasimov V, et al. Two clusters of human infection with influenza A/H5N1 virus in the Republic of Azerbaijan, February–March 2006. *Euro Surveill* 2006; **11:** 122–26.

20 WHO. Human cases of influenza at the human-animal interface, January 2014–April 2015. *Wkly Epidemiol Rec* 2015; **90:** 349–62.

21 Kayali G, Kandeil A, El-Shesheny R, et al. Active surveillance for avian influenza virus, Egypt, 2010–2012. *Emerg Infect Dis* 2014; **20:** 542–51.

22 Kandeel A, Manoncourt S, Abd el Kareem E, et al. Zoonotic transmission of avian influenza virus (H5N1), Egypt, 2006–2009. *Emerg Infect Dis* 2010; **16:** 1101–07.

23 Editorial team. Seven human cases of H5N1 infection confirmed in Azerbaijan, and one case in Egypt. *Euro Surveill* 2006; **11:** E060323.2.

24 WHO. Summary of human infection with highly pathogenic avian influenza A (H5N1) virus reported to WHO, January 2003–March 2009: cluster-associated cases. *Wkly Epidemiol Rec* 2010; **85:** 13–20.

25 WHO. Update on human cases of highly pathogenic avian influenza A(H5N1) virus infection, 2011. *Wkly Epidemiol Rec* 2012; **87:** 117–23.

26 WHO. Update on human cases of highly pathogenic avian influenza A(H5N1) virus infection, 2010. *Wkly Epidemiol Rec* 2011; **86:** 161–66.

27 WHO. Influenza at the human–animal interface: Summary and assessment as of 4 December 2014. http://www.who.int/influenza/human_animal_interface/Influenza_Summary_IRA_HA_interface_04December2014.pdf?ua=1 (accessed April 28, 2015).

28 WHO. Human cases of influenza at the human–animal interface, 2013. *Wkly Epidemiol Rec* 2014; **89:** 309–20.

29 Bowman AS, Nelson SW, Page SL, et al. Swine-to-human transmission of influenza A(H3N2) virus at agricultural fairs, Ohio, USA, 2012. *Emerg Infect Dis* 2014; **20:** 1472–80.

30 Cowling BJ, Jin L, Lau EH, et al. Comparative epidemiology of human infections with avian influenza A H7N9 and H5N1 viruses in China: a population-based study of laboratory-confirmed cases. *Lancet* 2013; **382:** 129–37.

31 Gao R, Cao B, Hu Y, et al. Human infection with a novel avian-origin influenza A (H7N9) virus. *N Engl J Med* 2013; **368:** 1888–97.

32 Uyeki TM, Cox NJ. Global concerns regarding novel influenza A (H7N9) virus infections. *N Engl J Med* 2013; **368:** 1862–64.

33 de Vries E, Guo H, Dai M, Rottier PJ, van Kuppeveld FJ, de Haan CA. Rapid Emergence of highly pathogenic avian influenza subtypes from a subtype H5N1 hemagglutinin variant. *Emerg Infect Dis* 2015; **21:** 842–46.

34 Baden LR, Kanapathipillai R, Campion EW, Morrissey S, Rubin EJ, Drazen JM. Ebola—an ongoing crisis. *N Engl J Med* 2014; **371:** 1458–59.

35 Cowling BJ, Yu H. Ebola: worldwide dissemination risk and response priorities. *Lancet* 2015; **385:** 7–9.

36 Zaki AM, van Boheemen S, Bestebroer TM, Osterhaus AD, Fouchier RA. Isolation of a novel coronavirus from a man with pneumonia in Saudi Arabia. *N Engl J Med* 2012; **367:** 1814–20.

37 WHO. Avian influenza A (H5N1) in Egypt update, 9 April 2015. http://www.emro.who.int/surveillance-forecasting-response/surveillance-news/avian-influenza-ah5n1-in-egypt-9-april-2015.html (accessed April 21, 2015).

38 WHO. Egypt: upsurge in H5N1 human and poultry cases but no change in transmission pattern of infection. May 15, 2015. http://www.emro.who.int/egy/egypt-news/upsurge-h5n1-human-poultry-cases-may-2015.html (accessed May 21, 2015).

39 WHO. Influenza at the human–animal interface: summary and assessment as of 31 March 2015. http://www.who.int/influenza/human_animal_interface/Influenza_Summary_IRA_HA_interface_31_March_2015.pdf?ua=1 (accessed April 28, 2015).

40 Arafa AS, Naguib MM, Luttermann C, et al. Emergence of a novel cluster of influenza A(H5N1) virus clade 2.2.1.2 with putative human health impact in Egypt, 2014/15. *Euro Surveill* 2015; **20:** 2–8.

41 WHO. Antigenic and genetic characteristics of zoonotic influenza viruses and development of candidate vaccine viruses for pandemic preparedness, September 2015. http://www.who.int/influenza/vaccines/virus/201509_zoonotic_vaccinevirusupdate.pdf?ua=1 (accessed Oct 7, 2015).

42 Uyeki TM. Human infection with highly pathogenic avian influenza A (H5N1) virus: review of clinical issues. *Clin Infect Dis* 2009; **49:** 279–90.

43 Mathur MB, Patel RB, Gould M, et al. Seasonal patterns in human A (H5N1) virus infection: analysis of global cases. *PLoS One* 2014; **9:** e106171.

44 Patel RB, Mathur MB, Gould M, et al. Demographic and clinical predictors of mortality from highly pathogenic avian influenza A (H5N1) virus infection: CART analysis of international cases. *PLoS One* 2014; **9:** e91630.

45 Uyeki TM. Global epidemiology of human infections with highly pathogenic avian influenza A (H5N1) viruses. *Respirology* 2008; **13** (suppl 1)**:** S2–S9.

46 Refaey S, Azziz-Baumgartner E, Amin MM, et al. Increased number of human cases of influenza virus A(H5N1) infection, Egypt, 2014–15. *Emerg Infect Dis* 2015; **21:** 2171–73.

47 Zhou L, Liao Q, Dong L, et al. Risk factors for human illness with avian influenza A (H5N1) virus infection in China. *J Infect Dis* 2009; **199:** 1726–34.

48 WHO. Global Alert and Response (GAR): Disease Outbreak News (DONs). http://www.who.int/csr/don/en/ (accessed April 28, 2015).

49 WHO. The Weekly Epidemiological Record (WER). http://www.who.int/wer/en/ (accessed April 28, 2015).

50 Western Pacific Region WHO. Avian Influenza Weekly Update. http://www.wpro.who.int/emerging_diseases/AvianInfluenza/en (accessed May 1, 2015).

51 FluTrackers. Global WHO and Ministries of Health confirmed H5N1 human cases list. https://flutrackers.com/forum/forum/flutrackers-high-pathogenic-h5n1-h1n08-h5n8-h5n6-h5n3-tracking-outbreaks-spread/720310-flutrackers-2015-global-who-ministries-of-health-confirmed-h5n1-human-cases-list (accessed April 28, 2015).

52 Moher D, Liberati A, Tetzlaff J, Altman DG, and the PRISMA Group. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. *BMJ* 2009; **339:** b2535.

53 WHO. Cumulative number of confirmed human cases of avian influenza A(H5N1) reported to WHO. http://www.who.int/influenza/human_animal_interface/H5N1_cumulative_table_archives/en/ (accessed April 28, 2015).

54 Centers for Disease Control and Prevention. Morbidity and Mortality Weekly Report. http://www.cdc.gov/mmwr/ (accessed April 28, 2015).

55 Mounts AW, Kwong H, Izurieta HS, et al. Case-control study of risk factors for avian influenza A (H5N1) disease, Hong Kong, 1997. *J Infect Dis* 1999; **180:** 505–08.

56 Centre for Health Protection. Hong Kong Special Administrative Region. Avian Influenza Report. http://www.chp.gov.hk/en/guideline1_year/29/134/332.html (accessed May 18, 2015).

57 WHO. WHO case definitions for human infections with influenza A(H5N1) virus, 29 August 2006. http://www.who.int/influenza/resources/documents/case_definition2006_08_29/en/index.html (accessed July 18, 2014).

58 WHO. Antigenic and genetic characteristics of zoonotic influenza viruses and candidate vaccine viruses developed for potential use in human vaccines. http://www.who.int/influenza/vaccines/virus/characteristics_virus_vaccines/en/ (accessed April 28, 2015).

59 Recombinomics. H5N1 clade 7 cases in China raise concerns, February 2009. http://www.recombinomics.com/News/02050903/H5N1_China_7_Concerns.html (accessed April 30, 2015).

60 Nguyen T, Rivailler P, Davis CT, et al. Evolution of highly pathogenic avian influenza (H5N1) virus populations in Vietnam between 2007 and 2010. *Virology* 2012; **432:** 405–16.

61 Smith GJ, Naipospos TS, Nguyen TD, et al. Evolution and adaptation of H5N1 influenza virus in avian and human hosts in Indonesia and Vietnam. *Virology* 2006; **350:** 258–68.

62 Wang J, Vijaykrishna D, Duan L, et al. Identification of the progenitors of Indonesian and Vietnamese avian influenza A (H5N1) viruses from southern China. *J Virol* 2008; **82:** 3405–14.

63 Le MT, Wertheim HF, Nguyen HD, et al. Influenza A H5N1 clade 2.3.4 virus with a different antiviral susceptibility profile replaced clade 1 virus in humans in northern Vietnam. *PLoS One* 2008; **3:** e3339.

64 Tian H, Cui Y, Dong L, et al. Spatial, temporal and genetic dynamics of highly pathogenic avian influenza A (H5N1) virus in China. *BMC Infect Dis* 2015; **15:** 54.

65 Gilbert M, Xiao X, Pfeiffer DU, et al. Mapping H5N1 highly pathogenic avian influenza risk in Southeast Asia. *Proc Natl Acad Sci USA* 2008; **105:** 4769–74.

66 Fang LQ, de Vlas SJ, Liang S, et al. Environmental factors contributing to the spread of H5N1 avian influenza in mainland China. *PLoS One* 2008; **3:** e2268.

67 Chaichoune K, Wiriyarat W, Thitithanyanont A, et al. Indigenous sources of 2007–2008 H5N1 avian influenza outbreaks in Thailand. *J Gen Virol* 2009; **90:** 216–22.

68 Rabinowitz PM, Galusha D, Vegso S, et al. Comparison of human and animal surveillance data for H5N1 influenza A in Egypt 2006–2011. *PLoS One* 2012; **7:** e43851.

69 Kuo HI, Lu CL, Tseng WC, Li HA. A spatiotemporal statistical model of the risk factors of human cases of H5N1 avian influenza in south-east Asian countries and China. *Public Health* 2009; **123:** 188–93.

70 Vong S, Ly S, Van Kerkhove MD, et al. Risk factors associated with subclinical human infection with avian influenza A (H5N1) virus—Cambodia, 2006. *J Infect Dis* 2009; **199:** 1744–52.

71 Yupiana Y, de Vlas SJ, Adnan NM, Richardus JH. Risk factors of poultry outbreaks and human cases of H5N1 avian influenza virus infection in West Java Province, Indonesia. *Int J Infect Dis* 2010; **14:** e800–05.

72 Lohiniva AL, Dueger E, Talaat M, et al. Poultry rearing and slaughtering practices in rural Egypt: an exploration of risk factors for H5N1 virus human transmission. *Influenza Other Respir Viruses* 2013; **7:** 1251–59.

73 Dinh PN, Long HT, Tien NT, et al. Risk factors for human infection with avian influenza A H5N1, Vietnam, 2004. *Emerg Infect Dis* 2006; **12:** 1841–47.

74 WHO. Influenza at the human–animal interface: summary and assessment as of 3 March 2015. http://www.who.int/influenza/human_animal_interface/Influenza_Summary_IRA_HA_interface_3_March_2015.pdf?ua=1 (accessed April 29, 2015).

75 Qin Y, Horby PW, Tsang TK, et al. Differences in the epidemiology of human cases of avian influenza A(H7N9) and A(H5N1) viruses infection. *Clin Infect Dis* 2015; **61:** 563–71.

76 Olsen SJ, Ungchusak K, Sovann L, et al. Family clustering of avian influenza A (H5N1). *Emerg Infect Dis* 2005; **11:** 1799–801.

77 Yang Y, Halloran ME, Sugimoto JD, Longini IM Jr. Detecting human-to-human transmission of avian influenza A (H5N1). *Emerg Infect Dis* 2007; **13:** 1348–53.

78 Wang H, Feng Z, Shu Y, et al. Probable limited person-to-person transmission of highly pathogenic avian influenza A (H5N1) virus in China. *Lancet* 2008; **371:** 1427–34.

79 Ungchusak K, Auewarakul P, Dowell SF, et al. Probable person-to-person transmission of avian influenza A (H5N1). *N Engl J Med* 2005; **352:** 333–40.

80 Webster RG, Govorkova EA. H5N1 influenza—continuing evolution and spread. *N Engl J Med* 2006; **355:** 2174–77.

81 Ibrahim M, Eladl AF, Sultan HA, et al. Antigenic analysis of H5N1 highly pathogenic avian influenza viruses circulating in Egypt (2006–2012). *Vet Microbiol* 2013; **167:** 651–61.

82 Reperant LA, Kuiken T, Osterhaus AD. Adaptive pathways of zoonotic influenza viruses: from exposure to establishment in humans. *Vaccine* 2012; **30:** 4419–34.

83 Monto AS. Vaccines and antiviral drugs in pandemic preparedness. *Emerg Infect Dis* 2006; **12:** 55–60.

84 WHO. Clinical management of human infection with avian influenza A (H5N1) virus. Aug 15, 2007. http://www.who.int/influenza/resources/documents/ClinicalManagement07.pdf?ua=1 (accessed Oct 7, 2015).

85 Li C, Bu Z, Chen H. Avian influenza vaccines against H5N1 'bird flu'. *Trends Biotechnol* 2014; **32:** 147–56.

http://dx.doi.org/10.1016/S1473-3099(16)00153-5

86 Swayne DE. Impact of vaccines and vaccination on global control of avian influenza. *Avian Dis* 2012; **56** (suppl): 818–28.

87 Swayne DE, Pavade G, Hamilton K, Vallat B, Miyagishima K. Assessment of national strategies for control of high-pathogenicity avian influenza and low-pathogenicity notifiable avian influenza in poultry, with emphasis on vaccines and vaccination. *Rev Sci Tech* 2011; **30:** 839–70.

88 WHO. Summary of status of development and availability of A(H5N1) candidate vaccine viruses and potency testing reagents. Sept 14, 2015. http://www.who.int/influenza/vaccines/virus/candidates_reagents/summary_a_h5n1_cvv_20150914.pdf?ua=1 (accessed Oct 7, 2015).

89 US Food and Drug Administration. FDA news release, 22 November 2013. http://www.fda.gov/NewsEvents/Newsroom/PressAnnouncements/ucm376444.htm (accessed April 21, 2015).

90 Adisasmito W, Chan PK, Lee N, et al. Effectiveness of antiviral treatment in human influenza A(H5N1) infections: analysis of a Global Patient Registry. *J Infect Dis* 2010; **202:** 1154–60.

91 Chan PK, Lee N, Zaman M, et al. Determinants of antiviral effectiveness in influenza virus A subtype H5N1. *J Infect Dis* 2012; **206:** 1359–66.

92 Govorkova EA, Baranovich T, Seiler P, et al. Antiviral resistance among highly pathogenic influenza A (H5N1) viruses isolated worldwide in 2002–2012 shows need for continued monitoring. *Antiviral Res* 2013; **98:** 297–304.

93 Trombetta C, Piccirella S, Perini D, Kistner O, Montomoli E. Emerging influenza strains in the last two decades: a threat of a new pandemic? *Vaccines (Basel)* 2015; **3:** 172–85.

94 Khuntirat BP, Yoon IK, Blair PJ, et al. Evidence for subclinical avian influenza virus infections among rural Thai villagers. *Clin Infect Dis* 2011; **53:** e107–16.

95 Gray GC, Krueger WS, Chum C, et al. Little evidence of subclinical avian influenza virus infections among rural villagers in Cambodia. *PLoS One* 2014; **9:** e97097.

96 Le MQ, Horby P, Fox A, et al. Subclinical avian influenza A(H5N1) virus infection in human, Vietnam. *Emerg Infect Dis* 2013; **19:** 1674–77.

97 WHO. WHO Comment on the importance of global monitoring of variant influenza viruses, 19 December 2011. http://www.who.int/influenza/human_animal_interface/avian_influenza/h5n1-2011_12_19/en/ (accessed Oct 7, 2015).

98 Fidler DP, Gostin LO. The WHO pandemic influenza preparedness framework: a milestone in global governance for health. *JAMA* 2011; **306:** 200–01.

99 WHO. Pandemic influenza risk management. WHO interim guidance. June 10, 2013. http://www.who.int/influenza/preparedness/pandemic/influenza_risk_management/en/ (accessed Oct 7, 2015).