

RESEARCH ARTICLES

Two clustered cases of confirmed influenza A(H7N9) virus infection, Beijing

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During April 1-8, 2017, a cluster of two laboratory-confirmed A(H7N9) influenza virus infections occurred in Yanqing district, Beijing. None of the 10 close contacts developed symptoms during the medical observation period after their last exposure. Throat swabs collected from close contacts all tested negative for influenza virus. Our study showed that the highly pathogenic avian influenza virus has spread to northern China, where the number of reported cases was relatively small, but the virus is not capable of sustained human-to-human transmission yet. In Beijing, informal live poultry trading in rural areas plays an important role in transmitting highly pathogenic H7N9 virus from poultry to human. Enhanced surveillance in ILI and SARI patients is required to strengthen the case detection capabilities during epidemic periods.

Introduction

In 2013, the first human case of avian influenza A(H7N9) virus infection was reported in China. By March 2019, over 1500 laboratory-confirmed human cases had been reported and the overall case fatality ratio was around 39% [1]. To prevent and control avian influenza infection and transmission, live poultry markets have been forbidden in Beijing since 2005 [2]. However, sporadic cases were still reported, and most cases had a history of exposure to poultry. In this study, we report the investigation of two clustered cases infected with A(H7N9) virus. We provide epidemiologic information on the outbreak, including history of poultry exposure, close contacts investigation, source of infection investigation and enhanced surveillance.

Methods*Patients*

Between April 1-8, 2017, a cluster of two laboratory-confirmed A(H7N9) influenza virus infections were reported in Yanqing district, Beijing. The two cases were brothers (aged 52 and 66) who had been exposed to poultry together and both developed symptoms on April 1.

Clinical and epidemiological collection

CDC staff collected clinical and epidemiological information by interviewing cases and their household members. The standard questionnaire provided in *The Protocol of Human Infection with Avian Influenza A(H7N9) virus Prevention and Control* was applied. Information included the demographics of cases, disease clinical course, history of poultry exposure within 10 days of illness onset, history of traveling and history of contact with persons with similar

symptoms. Close contacts and other exposed persons were identified. Close contacts were defined as individuals who contacted (< 1 meter) the patient without the use of personal protective equipment from the time of illness onset to the time when the H7N9 case was diagnosed and isolated.

Enhanced surveillance

As per the *Protocol of Human Infection with Avian Influenza A(H7N9) virus Prevention and Control*, enhanced surveillance was conducted in all secondary level and above medical institutions in the region in which the H7N9 cases were diagnosed. All patients that visited or were admitted to the sentinel hospitals were screened for influenza-like illness (ILI) symptoms and severe acute respiratory infection (SARI). Throat swabs were collected from ILI and SARI patients with their verbal consent and were tested for influenza virus.

Laboratory-testing

Throat swabs collected from cases were tested by PT-PCR according to the *Chinese Guideline of Diagnosis and Treatment for Human Infections with the Avian Influenza A (H7N9) Virus*, issued by the National Health and Family Planning Commission of China (NHFPCC)(3). Virus genetic sequences were obtained using Ion Torrent Personal Genome Machine sequencer (ThermoFisher, Guilford, CT, USA).

Results*Case descriptions*

Case 1 was a 66 year old man who lived in a village in Yanqing district, Beijing. On April 1, 2017, he experienced fever (39°C), chest pain, shortness of breath and cough with sputum. On April 4, he visited

a local hospital, as the disease had become more severe. Physical examination showed a body temperature of 40°C and a blood pressure of 170/90mmHg. Blood tests revealed a leukocyte count of $4.93 \times 10^9/L$ (neutrophils, 88.5%) and a C-reactive protein level of 114.25mg/L. Blood gas analysis revealed a pH of 7.5, a PaCO₂ of 22.8 mmHg and a PaO₂ of 53.0 mmHg. A chest computed tomography scan revealed a 'frosted glass' appearance (diffuse infiltrates) in both lungs. He was then diagnosed as a probable case with influenza A(H7N9) virus infection. On the evening of the same day, he was transferred to the intensive care unit of Ditan hospital. On April 5, a throat swab and sputum sample were collected from the patient and both samples tested positive for influenza A(H7N9) virus. On April 8, the 8th day of illness, the patient died.

Case 2 was the younger brother of case 1. He was a 52-year-old man. On April 1, he ran a fever (39.6°C) and developed respiratory distress, shortness of breath and fatigue. He visited a local community health service center and received symptomatic treatment. On April 4, day four of illness, he visited the infectious disease department of the district hospital. Physical examination showed a temperature of 38.3°C and a blood pressure of 111/88mmHg. Rales were not heard in the lungs. Blood tests revealed a leukocyte count of $5.85 \times 10^9/L$ (neutrophils, 63.4%). A chest X-ray revealed two increased lung markings and high-density shadows in left lung. A throat swab tested negative by rapid influenza detection test. The patient was administered Cefmetazole and aspirin-dl-lysine. The patient was diagnosed as probable case with A(H7N9) virus infection. On April 5, the patient was transferred to the intensive care unit of Ditan hospital. One day later, on April 6, the patient was diagnosed as laboratory-confirmed case of influenza A(H7N9) virus infection. On day 15 of illness, the patient was discharged from ICU. On April 26, the patient was discharged from hospital.

Epidemiologic Links and Exposure History

Between March 20 and April 4, case 1 stayed in his brother's (case 2) home in a village of Yanqing district, Beijing. Case 2 raised chickens in his backyard. On March 27, case 1 and case 2 bought over 70 chickens from a street peddler. Starting April 1, the newly bought chickens became sick and more than 10 of them died. Case 2 disposed of the dead chickens in a small brook nearby. Case 1 and case 2 had no other contact with live poultry or with patients with respiratory symptoms in the 10 days before illness onset. The street peddler was said to be from Hebei province and came to the town where the cases lived occasionally to sell poultry.

Close contact and co-exposed person investigation

A total of 9 close contacts were identified. Five were household contacts and four were neighbors of cases. They all had close contact with the cases but not with poultry. Another brother of the cases was identified as

a co-exposed person because he had fed the chickens. None of these 10 persons developed symptoms during the 10 day medical observation period after their last exposure. Throat swabs were collected from the 10 persons and all were negative for influenza virus.

Enhanced surveillance

From April 7 to April 20, a total of 173 ILI and 79 SARI patients were identified and 215 throat swabs were collected from them. All samples were tested for influenza virus by RT-PCR and one tested positive for influenza A(H7N9) virus (case 3). Case 3 was investigated and environmental samples from his home backyard were collected and tested.

Source of infection investigation

On April 6 and 7, 39 environmental samples were collected from the backyard of case 3. Of these, 11 tested positive for influenza A(H7N9) virus.

Genome sequence analysis demonstrated that the sequences from the virus isolated from case 1 were identical to those from case 2. The gene sequences of virus isolated from case 2 and the environment samples from case 3 showed that they shared 99.8% and 100.0% nucleic acid sequence. All isolated virus belonged to Yangtze River Delta evolution branch. The virus contained the Q226L substitution at the receptor binding sites in the HA, which is known to enhance the binding to mammalian α -2, 6-linked sialic acid receptors. Phylogenetic analysis in NA have not detected resistant mutations, and thus the virus should be sensitive to NA inhibitors.

Discussion

Since the first report of a human case of influenza A(H7N9) infection, human cases have spread across China, with most cases in Yangtze River Delta Region and Pearl River Delta Region (4). Sporadic human infections have been reported in Beijing, but less frequently than in South Eastern China. In this study, we report two clustered human cases caused by highly pathogenic H7N9 viruses.

Epidemiologic investigation showed that all three cases had contact with dead poultry before illness onset. Genome sequencing analysis showed that they were infected with highly pathogenic (HP) avian influenza virus. This is consistent with findings in other regions of China during the same period, which showed HP H7N9 virus emerged in poultry and caused human infections (5). Unlike H5N1, in many human cases of H7N9, there is no history of close contact with poultry (6). In February 2013, two human cases with HP A(H7N9) virus were identified in Guangdong, a city in southern China (7,8). Soon thereafter, additional human cases of HP virus infection were reported in other southern China cities (9). Our study shows that HP virus has spread to northern China, where the number of reported cases has been relatively small.

Through investigation, we found that the two clustered cases shared common exposure to poultry

and they developed symptoms on the same day as each other. Therefore, the infections were most likely caused via poultry exposure rather than human-to-human transmission. In addition, none of the close contacts got infected, suggesting the HP virus was not capable of sustained human-to-human transmission (10).

Previous studies have showed that live poultry trade plays an important role in H7N9 virus transmission, although much less than H5N1 (11). Although poultry markets have been banned in Beijing, informal live poultry trading continues (12). Poultry sellers move from wet market to streets and lanes in rural areas that are outside the scope of monitoring and surveillance. The epidemiological investigation and genome sequencing analysis of the isolated virus revealed that the two cases got infected from chickens bought from a street peddler. In addition, from interviews and genome analysis of samples from the case identified through enhanced surveillance (case 3), we found that case 3 had been exposed to dead chickens that were bought from a street peddler before illness onset and the genome sequences were highly similar with each other (99.95%-100%). This suggests that case 3 acquired his infection from the chickens he bought. Moreover, we found that there was 99.95%-100% similarity between viruses isolated from case 3 and case 2 (99.76%-100%). This suggests that the chickens might have been bought from the same street peddler, as case 3 and case 2 lived in different villages and they had no contact with each other. Our study demonstrates the role of illegal trading activities in transmitting viruses in Beijing, where formal LPMs have been completely prohibited. However, we failed to find the street peddler who transported and sold the chickens that carried H7N9 viruses. Most of them are cross-border sellers, whose activities are irregular. Our study demonstrates that illegal trade contributes to H7N9 virus infection in Beijing. According to a sampling investigation of adults in Beijing, 6.0% of them purchased live poultry within a year (13). This suggests that new approaches are necessary to manage informal live poultry trading in Beijing.

Although ILI and SARI surveillance are regularly performed in Beijing, their limited sampling rates might miss cases. Enhanced surveillance requires all ILI and SARI patients be sampled and tested with their consent, enhancing case detection capabilities. To conclude, the study reveals no evidence of human-to-human transmission of HP H7N9 virus in two related human cases. Illegal poultry trading played an important role in zoonotic H7N9 transmission.

References

1. World Health Organization(WHO). Influenza at the human-animal interface. Geneva: WHO, 2018 [2019-4-26]. Available from: https://www.who.int/influenza/human_animal_interface/Influenza_Summary_IRA_HA_inter face_02_03_2018.pdf?ua=1.
2. Li Q, Zhou L, Zhou M, et al. Epidemiology of Human Infections with Avian Influenza A(H7N9) Virus in China. *N Engl J Med*, 2014, 370(6):520-532. DOI: <https://doi.org/10.1056/NEJMoa1304617>
3. National Health and Family Planning Commission of the People's Republic of China. Chinese guideline of diagnosis and treatment for human infections with the avian influenza A(H7N9) virus (2014 edition). 2014-01-24.
4. Zhou L, Ren R, Yang L, et al. Sudden increase in human infection with avian influenza A(H7N9) virus in China, September-December 2016. *Western Pac Surveill Response J*, 2017,8(1):6-14. DOI: <https://doi.org/10.5365/wpsar.2017.8.1.001>
5. Zhang F, Bi Y, Wang J, et al. Human infections with recently-emerging highly pathogenic H7N9 avian influenza in China. *The Journal of infection*, 2017,75(1):71-75. DOI: <https://doi.org/10.1016/j.jinf.2017.04.001>
6. Bui C, Bethmont A, Chughtai AA, et al. A Systematic Review of the Comparative Epidemiology of Avian and Human Influenza A H5N1 and H7N9 - Lessons and Unanswered Questions. *Transboundary and Emerging Diseases*, 2015, 63(6):602-620. DOI: <https://doi.org/10.1111/tbed.12327>
7. European Centre for Disease Prevention and Control (ECDC). Genetic evolution of influenza A(H7N9) virus in China - implications for public health. Stockholm: ECDC, 2017 [2019-4-26] [http://ecdc.europa.eu/en/publications/Publications/09-mar-2017-RRA-update-6-influenza-A-\(H7N9\)-China-Taiwan.pdf](http://ecdc.europa.eu/en/publications/Publications/09-mar-2017-RRA-update-6-influenza-A-(H7N9)-China-Taiwan.pdf).
8. World Health Organization (WHO). Human infection with avian influenza A(H7N9) virus - China. Geneva: WHO, 2017. [2019-4-26]. Available from: <http://www.who.int/csr/don/27-february-2017-ah7n9-china/en/>.
9. Zhou L, Tan Y, Kang M, et al. Preliminary Epidemiology of Human Infections with Highly Pathogenic Avian Influenza A(H7N9) Virus, China, 2017. *Emerging Infectious Diseases*, 2017, 23(8):1355-1359. DOI: <https://doi.org/10.3201/eid2308.170640>
10. European Centre for Disease Prevention and Control (ECDC). Highly pathogenic A(H7N9) virus mutation does not change risk to humans: Although the genetic changes in A(H7N9) may have implications for poultry, to date, there is no evidence of increased transmissibility to humans or sustainable human-to-human transmission. *ScienceDaily*. ScienceDaily, 2017-03-13 [2019-4-16]. Available from: <http://www.sciencedaily.com/releases/2017/03/170313102354.htm>.
11. Bethmont A, Bui CM, Gardner L, et al. Quantified degree of poultry exposure differs for human cases of avian influenza H5N1 and H7N9.

- Epidemiology and Infection, 2016, 144(12):2633-2640. DOI: <https://doi.org/10.1017/S0950268816001035>
12. Fournie G , Pfeiffer D . Can closure of live poultry markets halt the spread of H7N9?. *Lancet*, 2014, 383(9916):496-497. DOI: [https://doi.org/10.1016/S0140-6736\(13\)62109-1](https://doi.org/10.1016/S0140-6736(13)62109-1)
 13. Wu S, Yang P, Wang Q, et al. Human exposure to live poultry among residents during the second wave of avian influenza A (H7N9) epidemic in Beijing, 2013-2014. *Chinese Journal of Epidemiology*, 2017, 38(11):1484.

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