Clinical features of three avian influenza H7N9 virus-infected patients in Shanghai

Xiao Fei Wang, Guo Chao Shi, Huan Ying Wan, Shao Guang Hang, Hong Chen, Wei Chen, Hong Ping Qu, Bao Hui Han* and Min Zhou

Department of Respiratory Medicine, School of Medicine, Ruijin Hospital, Shanghai Jiao Tong University, Shanghai, China

Abstract

Introduction: Since February 2013, a novel reassortant H7N9 virus associated with human deaths, but no apparent outbreaks in poultry and wild birds has emerged in eastern China.

Objectives: The potential reemergence of H7N9 during next year's influenza season demand a further understanding of this important disease.

Methods: Between March 1 and April 30, 2013, we obtained and analyzed clinical, epidemiologic and radiologic features, and virologic data from three laboratory-confirmed patients of A H7N9 infection admitted in Shanghai Ruijin Hospital.

Results: All patients were middle to old aged (mean age 62 years) and overweight (mean body mass index 31) patients. Two patients were exposed to poultry directly or indirectly in food market. They presented with fever and rapidly progressive pneumonia that did not respond to antibiotics. Time between onset of symptoms and onset of respiratory failure (days) were 7–11 days. Two patients presented secondary invasive bacterial infections. All patients died on day 7 to day 86 after the onset of symptoms.

Conclusions: Cross species poultry-to-person transmission of this new reassortant avian influenza H7N9 virus can result in severe and fatal respiratory disease like acute respiratory distress syndrome (ARDS) in humans. Reduplicate chest imaging examination is suggested for risky patients with fever and dyspnea. Secondary invasive bacterial infections and pneumothorax can cause severe and fatal consequence. Old age, obesity and presence of comorbidity may be associated with increased mortality. Pulmonary fibrosis can be seen at late stage of the disease.

Please cite this paper as: Wang XF, Shi GC, Wan HY, Hang SG, Chen H, Chen W, Qu HP, Han BH and Zhou M. Clinical features of three avian influenza H7N9 virus-infected patients in Shanghai. *Clin Respir J* 2014; 8: 410–416.

Authorship and contributorship

Xiao Fei Wang, Guo Chao Shi, Huan Ying Wan, Shao Guang Hang, Hong Chen, Wei Chen, Hong Ping Qu were responsible for data collection. Xiao Fei Wang, Bao Hui Han were responsible for manuscript writing and editing. All authors have seen and approved the final version of the manuscript.

Ethics

The study protocol was approved by the Coordinating Ethics Committee of Ruijin Hospital.

Conflict of interest

The authors have stated explicitly that there are no conflicts of interest in connection with this article.

*Co-correspondence author: Shanghai Chest Hospital, Shanghai Jiao Tong University.

Influenza A subtype H7 virus is a recognized common virus that can cause infection in poultry, which occurs worldwide. However, Mainland China has just experienced the first outbreak of novel reassortant avian-origin influenza H7N9 virus infection in humans (1).

We obtained and analyzed the clinical, epidemiologic, laboratory and radiologic features from three patients admitted in Ruijin Hospital. Respiratory specimens were tested for influenza by means of real-time reverse transcriptase–polymerase chain reaction (rt-PCR) assays and confirmed to be H7N9 positive.

Case history

Case 1

The first case occurred in a 67-year-old obese woman with hypertension and diabetes who had traveled to
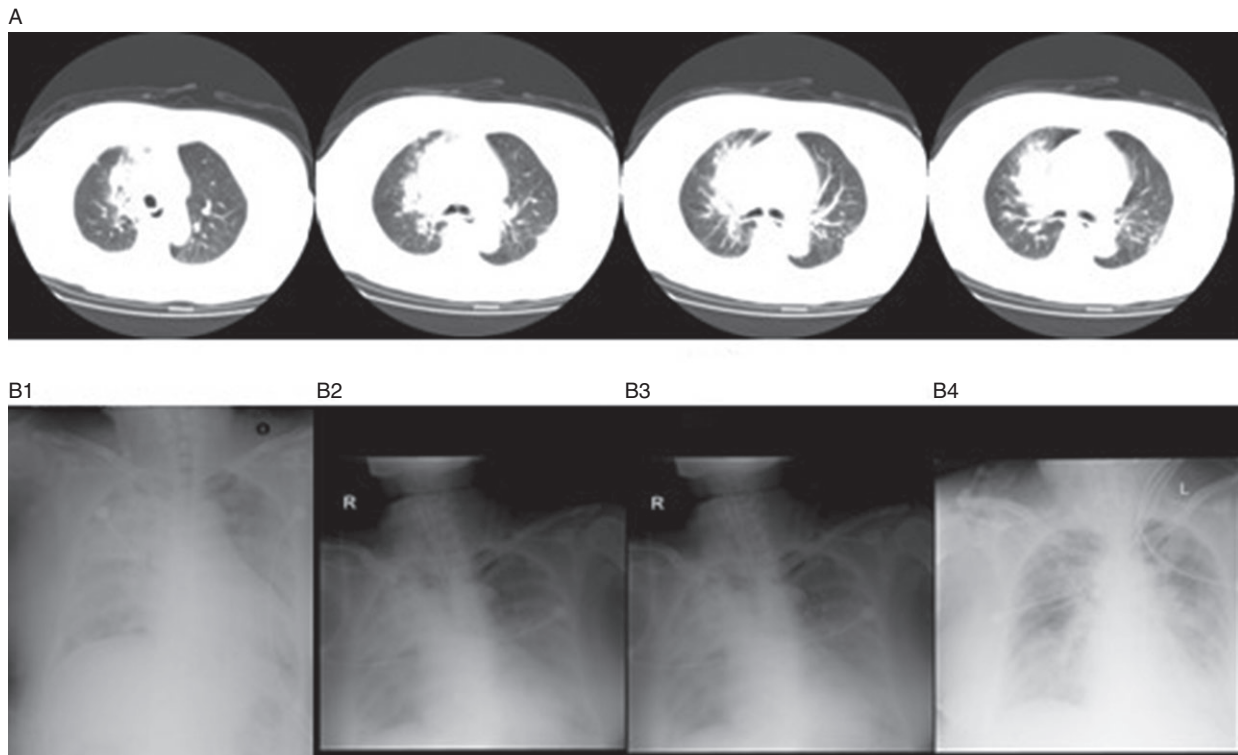
A

B1 B2 B3 B4

Figure 1. Chest imaging of case 1. (A) Computed tomography scan on d9 of symptoms; (B1) X-ray on d7 of symptoms; (B2), X-ray on d14 of symptoms; (B3) X-ray on d15 of symptoms; (B4) X-ray on d20 of symptoms.

Zhe Jiang Province with the family a week before hospitalization. No direct exposure to poultry was noticed, and nobody else in the family was sick. Fever and headache developed in this patient just after her return. She was admitted to Shanghai Orient Hospital on March 25. On admission, she had oral temperature of 38.7°C. The chest was clear to auscultation, and the blood test was normal. The patient presented no amelioration in spite of intravenous administration of vidarabine and cefotiam with standard dose, so she came to our emergency on March 27. Computed tomography scan showed consolidation in the right upper lobe (Fig. 1A) while total white cell count (5.35×10^9/L) and neutrophiles (3.65×10^9/L) remained normal, but serum creatine kinase increased (CK 582 IU/L). Intravenous ciprofloxacin 400 mg once a day was added to the regimen. However, the gas analysis realized on March 29 on room air presented a severe hypoxia (PaO_2 : 6.05kPa), and her pulse oxygen saturation fluctuated between 78% and 82% despite the use of biphasic positive airway pressure (BiPAP). So the patient was transferred to specialist intensive care unit (ICU) on March 31, where the intubation was done. Specimens obtained from nose and throat swabs by the Centers for Disease Control and Prevention (CDC) on

day 2 of hospitalization were confirmed to be positive for H7N9 by means of rt-PCR. The patient stayed 14 days in the ICU; pulmonary infiltrations rapidly extended to both lungs (Fig. 1B), and clinically she continued worsening despite the antiviral and antibiotic treatment (oseltamivir 150 mg twice a day through nasogastric tube + imipenem 500 mg q6h iv + linezolid 600 mg q12h iv). The patient finally died on April 13. Microbiology laboratory reported that the blood culture of this patient was positive for *Burkholderia cepacia* 3 days after her death.

Case 2

Patient 2 was a 64-year-old male who weighs 90 kg, with a body mass index (BMI) of 33. He had 10 years' history of hypertension and 5 years' history of diabetes. He went to the emergency of our hospital on April 3 for persisted fever to 39°C despite the use of paracetamol. The blood test was normal (white cell count 5.62×10^9/L, lymphocyte count 3.43×10^9/L, platelet count 94×10^9/L) with increased serum CK level (CK 537IU/L); the X-ray showed patchy consolidation of upper lobe of her right lung (Fig. 2A). Intravenous levofloxacin, 500 mg once a day + cefoxitin, 2 g
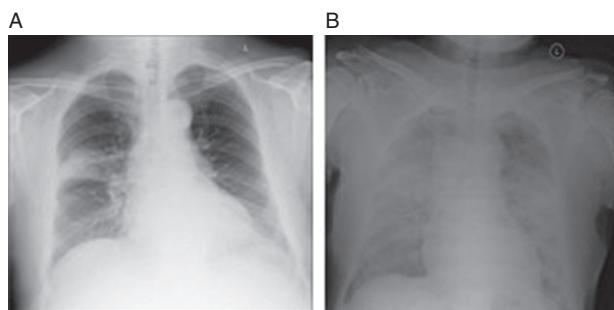
Figure 2. Chest radiographs of case 2 patient. (A) X-ray on d3 of symptoms; (B) X-ray on d7 of symptoms.

twice a day were prescripted, however the patient showed up at the emergency again on April 7 for breathlessness. Auscultation revealed fine moist in the lower lung fields. Sequential chest X-rays showed extensive involvement of the right lung, with new infiltrates appearing on the left (Fig. 2B). The arterial gas analysis showed a severe hypoxia (PaO_2 5.47 kPa, SaO_2 61%) on room air, requiring noninvasive ventilation with BiPAP. We noticed the elevation of C-reactive protein (73 mg/L) and CK (537 IU/L) at the same time. After throat swab samples were taken, oral oseltamivir (150 mg twice a day) was immediately added to the regimen. However, the patient showed no amelioration with BiPAP, and unfortunately he died in the course of intubation. Reports from CDC confirmed this patient to be H7N9 positive; interrogation of the family revealed that the patient went to the food market every day when he was healthy, which is believed to be the source of infection for this patient.

Case 3

Patient 3 was a 54-year-old man with no underlying health problems. He had bought a killed chicken in the food market 5 days before the sickness. He came to our emergency on April 9 for persisting high fever after 3 days' antivial treatment (ribavirin). The first X-ray (Fig. 3A) was taken in emergency and showed consolidation in right middle lobe; 1 day later, the lesions extended rapidly to right lower lobe and left middle lobe (Fig. 3B). Like the first two cases, the hemogram was normal whereas CK increased to 2758 IU/L and PaO_2 decreased to 5.13 kPa. The patient was intubated on April 10, and he was laboratory confirmed to be the third case of A H7N9 virus infection on the same day. Two weeks later, he was successfully extubated and now sequentially supported by BiPAP Vision (Respironics, Murrysville, PA, USA) with stable vital signs. The rt-PCR result of his throat swabs turned negative of H7N9 after 21 days' use of oseltamivir (150 mg twice a day through nasogastric tube from April 10 to April 30). Repeated sputum cultures of the patient were positive for *Stenotrophomonas maltophilia* since April 27, but the blood cultures remained sterile. The patient became apyrexic on May 4 with antibiotherapy of cefepime (2 g q12h iv). Generalized coarse reticulation of the right lung was noted on the chest radiograph realized on May 5 (Fig. 3C), which may explain why the patient still rely on BiPAP support with high concentration of oxgen therapy (fraction of inspiration oxygen 80%). The patients had been relatively stable with BiPAP until May 18, when his dyspnea gradually worsened. Bedside chest radiograph (Fig. 3D) showed right-sided pneumothorax as was suspected, and an intercostal chest drain was inserted the same day. The patient suffered of refractory
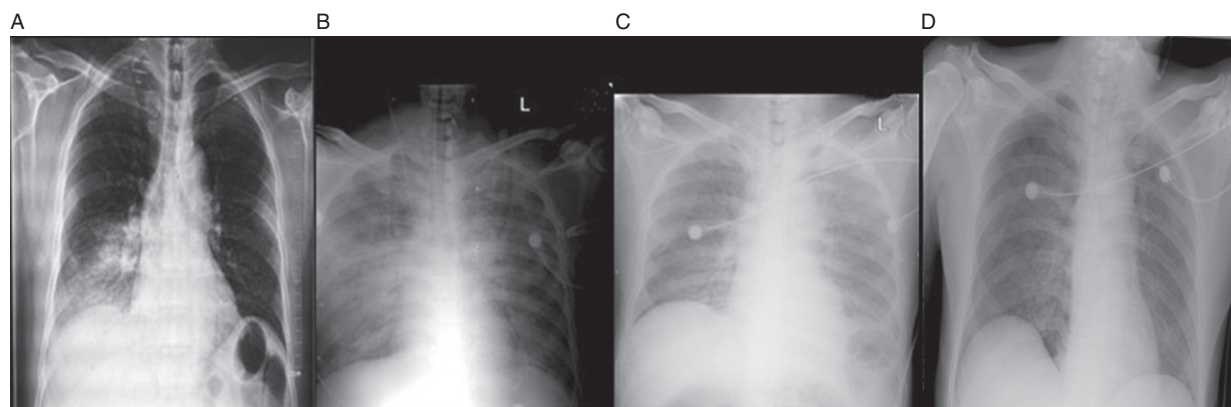

Figure 3. Chest radiographs of case 3. (A) X-ray on d3 of symptoms; (B) X-ray on d4 of symptoms; (C) X-ray on d29 of symptoms; (D) X-ray on d42 of symptoms.

Table 1. Demographic, epidemiologic, and imaging characteristics and treatment and clinical outcome of three patients infected with A H7N9 virus

Characteristic	Case 1	Case 2	Case 3
Age (year)	67	64	54
Sex	F	M	M
Occupation	Retired	Retired	Retired
BMI	35	33	26
Underlying illness	Hypertension, diabetes	Hypertension, diabetes	None
Exposure to live poultry	None	Yes (probably indirect)	Yes
Time between onset of symptoms and initiation of oseltamivir (days)	14	7	8
Time between onset of symptoms and onset of respiratory failure (days)	11	7	8
Time between onset of respiratory failure and need for mechanical ventilation (days)	3	1	1
Time between mechanical ventilation and death (days)	14	1	78
Major symptom	Fever, dyspnea	Fever, dyspnea	Fever, dyspnea
Hemogram at presentation	Normal	Normal	Normal
CK (IU/L) at presentation	582	537	2758
Initial affected lobe on X-ray	Upper lobe of right lung	Upper lobe of right lung	Middle lobe of right lung
Complications	Sepsis shock (*Burkholderia cepacia*)	Unknown	Secondary bacterial pneumonia (*Stenotrophomonas maltophilia*) and pneumothorax
Antibiotics given	Cefotiam, ciprofloxacin, imipenem–cilastatin, vancomycin, moxifloxacin, linezolid, caspofungin	Levofloxacin, cefoxitin, imipenem–cilastatin	Cefuroxime, cefotaxime, cefepime, vancomycin, meropenem
Days after onset of symptoms on which intravenous methylprednisolone given (dosage)	Days 11–23 (40 mg every 24 h days 11–13; 40 mg every 8 h days 14–17; 40 mg every 24 h days 18–23)	Day 7 (40 mg every 12 h)	Days 6–26 (40 mg every 24 h days 6–9; 40 mg every 8 h days 10–12; 40 mg every 12 h days 13–15; 40 mg every 24 h days 15–20; 20 mg every 24 h days 21–26)
Days after onset of symptoms on which intravenous immunoglobulin given (dosage)	Days 17–25 (10 g every 24 h)	Day 7 (5 g every 24 h)	Days 10–26 (20 g every 24 h)
Outcome	Died on April 13 of sepsis choc	Died on April 7 in course of intubation	Died on June 26 of refractory pneumothorax

BMI, body mass index; CK, creatine kinase.

hypoxia because of the persisting pneumothorax and finally died on June 26.

Results

We identified three patients with mean age of 62 years including two females and one male (the clinical characteristics of these three patients are shown in Table 1). Two patients were exposed to live poultry directly or indirectly in the food market while the other patient got a trip outside Shanghai; the presumed incubation period was about 1 week. All of the patients were overweight to obese, with BMI ranging from 26 to 35 (mean 31). None of the patients had upper respiratory tract symptoms or conjunctivitis. Fever and later onset dyspnea were the major symptoms. The

Table 2. Summary of human infection with avian flu in recent years

Subtype of virus	Source	Year/country	Number/clinical manifestation
A/H7N7	Gull	1980/United States	3/conjunctivitis
A/H7N7	Duck	1996/United Kingdom	1/conjunctivitis
A/H5N1	Bird	1997/Hong Kong, China	18/ILI, pneumonia
A/H9N2	Chicken	1998/Guang Dong, China	5/ILI, pneumonia
A/H9N2	Avian	1999/Hong Kong, China	2/ILI
A/H7N2	Avian	2002/North America (Virginia)	1/ILI, serologic diagnostic
A/H9N2	Avian	2003/Hong Kong, China	1/ILI
A/H5N1	Avian	2003/Hong Kong, China	2/ILI
A/H7N7	Avian	2003/Holland	89/conjunctivitis, ILI, pneumonia (including 1 death)
A/H5N1	Avian	2003 until now, 15 countries	602/ILI, pneumonia
A/H7N2(NY/107)	Unknown	2003/North America (New York)	1/pneumonia
A/H7N3	Avian	2004/Canada	2/conjunctivitis, ILI; 1 LPAI; 1 HPAI
A/H5N2	Avian	2005/Japan (Ibaraki)	13/serologic diagnostic
A/H7N2	Avian	2007/United Kingdom (Wales)	1/conjunctivitis, ILI
A/H9N2	Unknown	2007/Hong Kong, China	1/ILI
A/H10N7	Duck	2004/Eygept (Ismaillia)	2/ILI
A/H10N7	Birds from infected areas	2010/Sydney (New South Wales)	7/URI, conjunctivitis

HPAI, highly pathogenic avian influenza; ILI, influenza-like infection; LPAI, lowly pathogenic avian influenza; URI, upper respiratory tract infection.

hemograms were normal at presentation, and we observed a substantial elevation of CK in all three patients. Upper and middle lobe of the right lung are the most affected zones at early stage of the disease, which may be related to the inhalation of the virus by respiratory tract, and then rapidly extended to both lungs within 7–15 days (mean 10 days). Antiviral therapy with traditional medications like vidarabine and ribavirin showed no effect on this kind of infection. Three patients were given 150 mg oral oseltamivir twice daily after throat swab samples were taken, starting a mean of 9.7 days after onset of symptoms. Mean time between onset of symptoms and respiratory failure was 8.6 days. All patients required mechanical ventilation. Combination antibiotic therapy, glucocorticoids and intravenous immunoglobulin were administered in all three patients. Patient 1 died of sepsis 14 days after intubation while patient 2 died of cardiac arrest in course of intubation. The third patient had once been successfully extubated; however, he later complicated with a pneumothorax in the context of pulmonary fibrosis and finally died 86 days after the onset of symptoms. Secondary invasive bacterial infections were found in two of these patients: sepsis choc of *B. cepacia* in patient 1 and bacterial pneumonia of *S. maltophilia* in patent 3.

Discussion

Human infections with avian-origin influenza virus have been observed before (Table 2) (2); most infec-tions are generally mild, causing conjunctivitis or modest respiratory symptoms, although a fatal case was reported before this A H7N9 virus outbreak (3).

Novel influenza viruses of the A H7N9 subtype, characterized by high fever and severe respiratory symptoms, have infected 127 and killed 24 people in China as of April 30, 2013 and may pose a serious human health risk. An understanding of the source and mode of transmission of these infections, further surveillance and appropriate countermeasures are urgently required. This novel influenza viruses' hemagglutinin (HA) and neuraminidase genes probably originated from Eurasian avian influenza viruses; the remaining genes are closely related to avian H9N2 influenza viruses. Several characteristic amino acid changes in HA and the polymerase basic 2 protein (PB2) RNA polymerase subunit probably facilitate binding to human-type receptors and efficient replication in mammals, respectively, highlighting the pandemic potential of the novel viruses (4) (Fig. 4). However, unlike other types of avian influenza affecting human beings, no increase in poultry deaths was noticed before onset of human infections. Zoonotic infections of A H7N9 virus from birds to humans appear to be associated with live poultry markets, like the last two of our cases. There also have been sporadic reports of human-to-human transmission (5, 6); therefore, the pandemic potential of these novel avian-origin viruses should not be underestimated. We diagnosed avian influenza H7N9 in all three patients (who were epidemiologically unlinked); two patients had
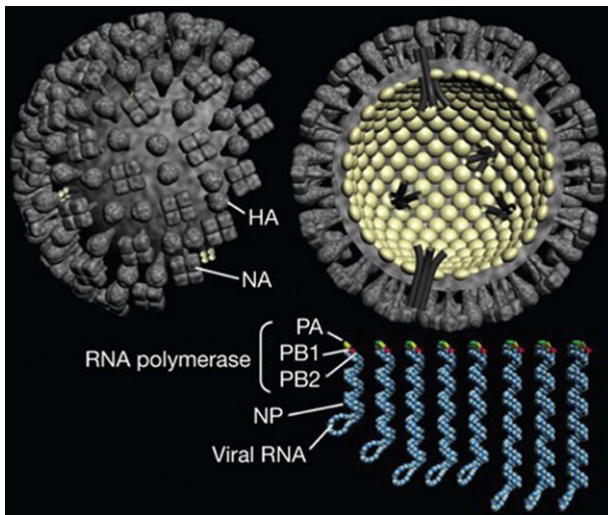
Figure 4. Structure of avian influenza virus H7N9.

histories of wet market exposure to poultry, suggesting poultry-to-person transmission. Fortunately, no close contacts of these three patients are found be infected till now.

Our patients presented high fevers, lower respiratory tract symptoms (especially dyspnea) and radiological features of consolidation without upper respiratory tract symptoms. The most striking common preexisting factor of our patient group was obesity or being overweight, which was previously reported to be a risk factor for developing acute respiratory distress syndrome (ARDS) in H1N1 infection (7, 8). In another study, development of ARDS was seen more frequently in obese patients (9). However, in a large cases study (10), they did not find that obesity was associated with increased mortality. It is may be because our patients are more severe and presented ARDS. The first two patients who died within 1 month were both over 60 and associated with underlying health problems (hypertension and diabetes). The third patient, the 54-year-old male with no medical history, has once been successfully extubated. Thus, we think that poor prognosis may probably be related with old age (>60 years), obesity and presence of comorbidity. All three patients received treatment with oseltamivir starting on day 7–14 of symptoms onsets, probably at the time of respiratory failure onset. Data related to human infections with seasonal, pandemic and HPAI H5N1 viruses indicate that the earlier antiviral treatment is initiated, the greater the clinical benefit (11). However, none of these clinical, radiological or laboratory changes was pathognomonic, which result in the delay of the antiviral treatment. So this reminds us to pay attention to 'risky' patients with copresence with of fever and dyspnea with normal white blood cell count and CK elevation, especially in those old and overweight patients.

Virus is not the only germ we face; secondary invasive bacterial infections were found in two of our patients, and one result in fatal sepsis. The development of secondary infection, especially Gram-negative rod infection, can be explained by several factors, such as exposure to high-risk procedures, use of glucocorticoids, severity of underlying diseases and frequent use of medical instruments, all of which may facilitate development of nosocomial bacteremia (12), so appropriate empirical antibiotic treatment may be indicated for initial management of severe A H7N9 virus pneumonia. Glucocorticoids are used in all of our cases while they are still controversial in the routine treatment of influenza (13–15). Because of the limited number of patients, our experiences are not conclusive; whether the continuous activity of virus replication is associated with glucocorticoid is unknown. Long-term complication of mechanical ventilation is pneumothorax. Yang and her colleagues (16) find that the incidence of pneumothorax in serious coronavirus [severe acute respiratory syndrome (SARS)-CoV] patients with noninvasive positive pressure ventilation is significantly higher than that without receiving MV. This might be related to viral-related pulmonary injuries, intensive cough and high mechanical ventilation pressure.

The outcome of A H7N9 virus infection in human beings is unfavorable, especially in cases present with ARDS. From the imaging evolution of the only survivor, we find that the opacification has largely resolved and replaced by reticulations especially in the initially affected lung. The histologic evolution of SARS-CoV infection coincided with the different stages of diffuse alveolar damage: acute, proliferative organizing and fibrotic stages (17); whether the nrH7N9 infection shared the same process is unknown.

Concerning the high mortality of these infections, early diagnosis is the key for the treatment. Reduplicate chest imaging examination is suggested for risky patients with fever and dyspnea; early use of Oseltamivir is necessary for suspicion patients. Old age, obesity and presence of comorbidity may be associated with increased mortality.

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