CHAPTER I

INTRODUCTION

1.1 Background and significance of the problem

Influenza, commonly known as flu, is an infectious disease of birds and mammals caused by RNA viruses of the Orthomyxoviridae (the influenza viruses) family. There are three genera of influenza virus: Influenza virus A, Influenza virus B and Influenza virus C. Each genus includes only one species, or type: Influenza A virus, Influenza B virus, and Influenza C virus, respectively. Influenza A and C infect multiple species, while influenza B almost exclusively infects human (Hay et al., 2001). Influenza A viruses are further sub classified, based on the viral surface proteins hemagglutinin (HA or H) and neuraminidase (NA or N). Sixteen H subtypes (or serotypes) and nine N subtypes of influenza A virus have been identified. The type A viruses are the most virulent human pathogens among the three influenza types and cause the most severe disease. The Influenza A virus subtypes are labeled according to an H number (for hemagglutinin) and an N number (for neuraminidase). Each subtype virus has mutated into a variety of strains with differing pathogenic profiles; some pathogenic to one species but not others, some pathogenic to multiple species (Atkinson et al. 2007). Influenza A virus subtype H5N1, also known as A(H5N1) or simply H5N1, is a subtype of the Influenza A virus which can cause illness in human and many other animal species.

A bird-adapted strain of H5N1, called HPAI A (H5N1) for "highly pathogenic avian influenza virus of type A of subtype H5N1", is the causative agent of H5N1 flu, commonly known as "avian influenza" or "bird flu". It is endemic in many bird populations, especially in Southeast Asia (Webster et al., 2006). Sporadic transmission to human raises concern that the H5N1 virus may mutate to generate a novel strain capable of sustained human-to-human transmission with pandemic potential. The World Health Organization has described the threat from H5N1 as a "public health crisis", and declared that the world is at increasing risk of an avian influenza A (H5N1) pandemic.(Chutinimitkul et al., 2006)

Avian influenza A (H5N1) epidemic situation in the world

From the end of 2003 to date, avian influenza A (H5N1) virus infection has been confirmed in animals and/or humans in several countries in Asia, Europe, and Africa. There is rapid geographical spread of the virus, and increasing potential for human infection. One special concern is the potential for human-to-human transmission. So far, the spread of H5N1 virus from person to person has been rare. In mid-2003, the highly pathogenic H5N1 virus began to circulate widely in poultry in parts of south-east Asia, spreading within months to affect 8 countries in an outbreak unprecedented in its geographic extent (Figure 1).
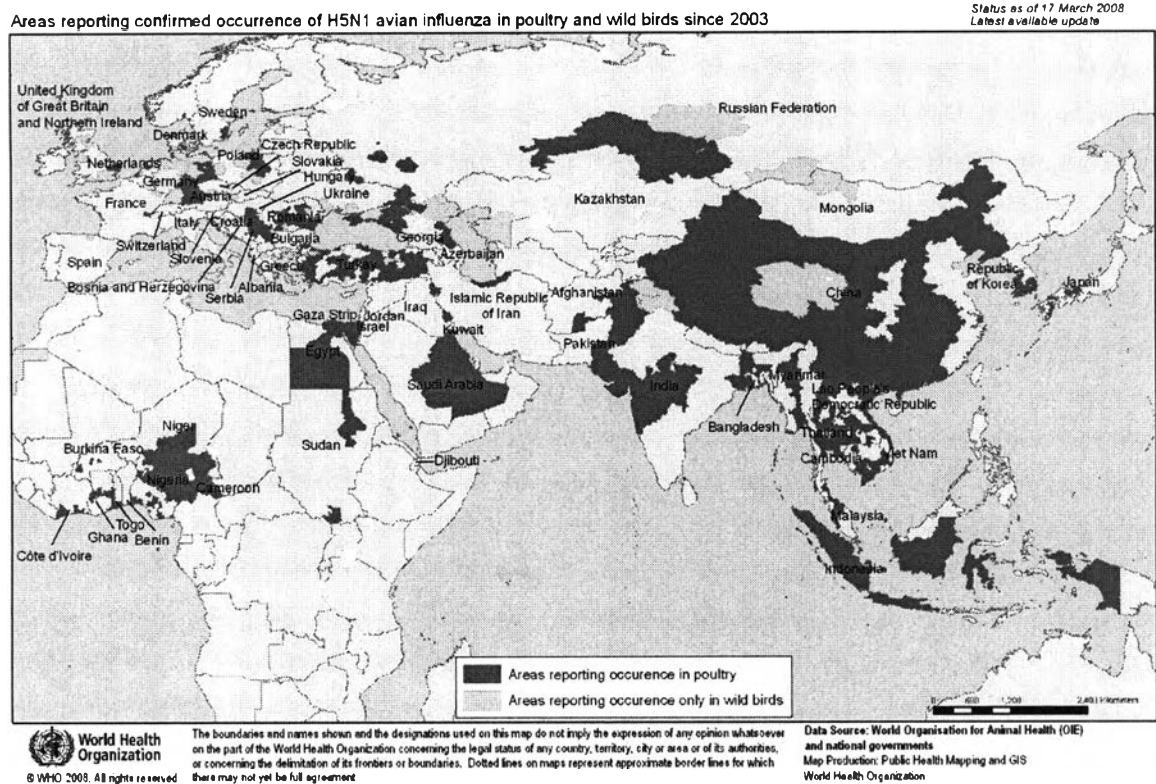
Areas reporting confirmed occurrence of H5N1 avian influenza in poultry and wild birds since 2003

Figure 1: Map of aresa reporting confirmed occurrence of avian influenza (H5N1)

in poultry and wild birds since 2003. (WHO, 17 March 2007)

In parallel with the large outbreaks of avian influenza A (H5N1), human cases

of H5N1 infection also occurred. The first human H5N1 outbreak occurred in Hong

Kong in 1997, which caused not only widespread poultry deaths, but also 18 human

cases with six deaths (Chan, 2002). From December 1 2003 to March 18 2008, 14

countries reported a total of 373 laboratory-confirmed human cases of avian influenza

A (H5N1) to World Health Organization (WHO)(World Health Organization [WHO],

2007). More than half of all cases were reported by Vietnam (106 cases) and
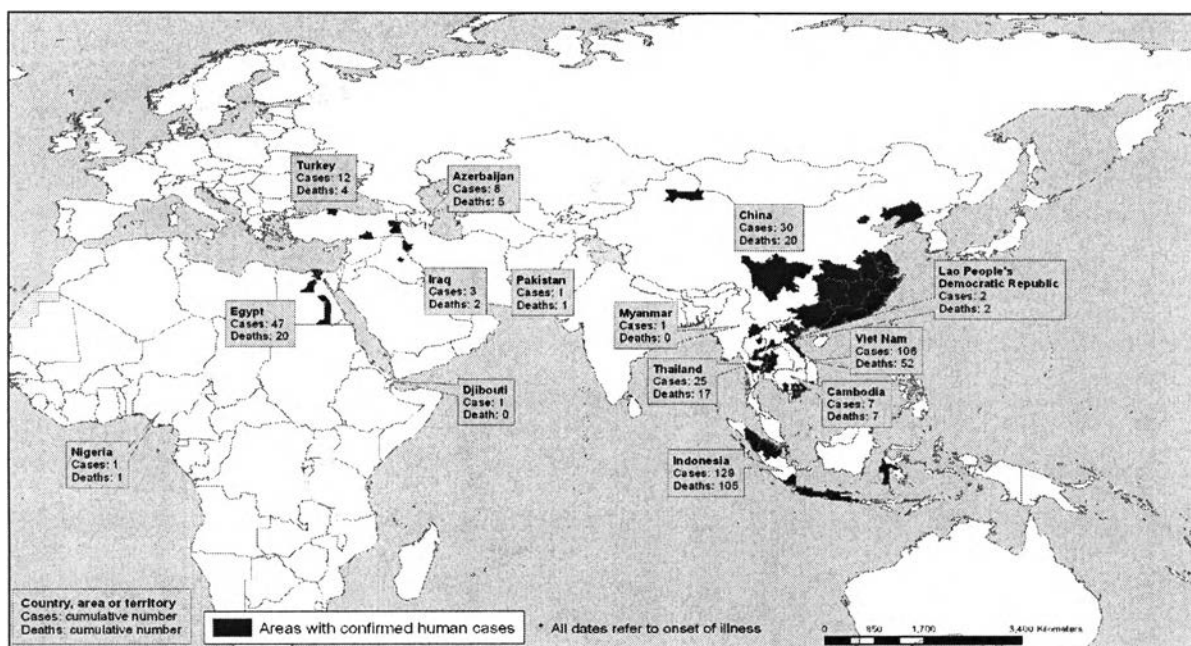
Indonesia (129 cases) (Figure 2).

Figure 2: Map of areas with confirmed human cases of avian influenza

A (H5N1) since 2003 (WHO, 18 March 2008)

Avian influenza A (H5N1) in China

In China, the most recent reported outbreak occurred in a wet poultry market in Guangzhou of Guangdong province. At least 144 chickens have since been reported dead. As of March 13, 2008 (OIE, 2007). In 2004, there were a total 50 outbreaks of poultry H5N1 outbreaks in 16 provinces in China, but no confirmed human H5N1 cases were identified. Surveillance for communicable diseases is the main public health surveillance activity.

In China, all suspected H5N1 cases are reported to the Chinese Center for Disease Control and Prevention (China CDC) through a national surveillance system. On October 12, 2005, the first confirmed human case of avian influenza A (H5N1) in China was detected by China CDC (Yu et al., 2006). From October 2005 through

March 2008, Of the 28 laboratory-confirmed civilian cases distributed widely in 13 provinces in China, 18 have been fatal (WHO, 2007). The count does not include two military cases reported in 2003 and 2007 (Figure 3). Only 4 of 23 cases were temporally associated with confirmed H5N1 poultry outbreaks. (Figure 4)

Figure 3: Confirmed cases of human avian influenza A (H5N1),

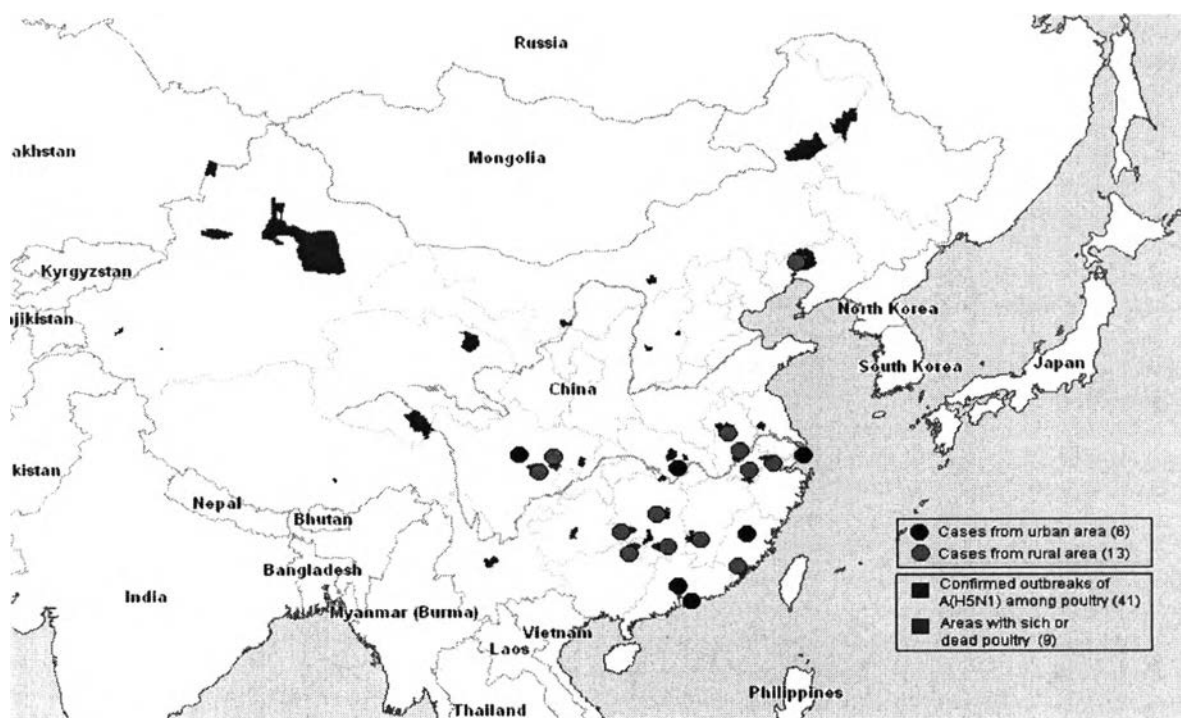by date of onset - China, 2005 - 2008

Figure 4: Map of confirmed cases of human avian influenza A (H5N1),

China, 2005 – 2007

Field investigations will perform to determine key epidemiological and clinical characteristics for cases. However, because collection, shipment, and testing of specimens often require several days or longer, the investigation will begin before laboratory test results are available for persons with pneumonia of unknown origin (PUO) suspected of having H5N1 infection. Clinical specimens obtained from suspect cases will be tested in laboratory of provincial level by conventional reverse transcriptase polymerase chain reaction (RT-PCR) and real time reverse transcriptase PCR assay. PCR can be targeted at genes that are relatively conserved across all influenza A viruses. Once Influenza/HA confirmed, specimens will be transferred to National Influenza Center (NIC) for serological tests and viruses culture. The national health authority will notify WHO and mass media of confirmed human case of H5N1 right after the disease was confirmed by NIC. The process and conceptual framework

of study show as following figure and details will be discussed in chapter III (Figure5).
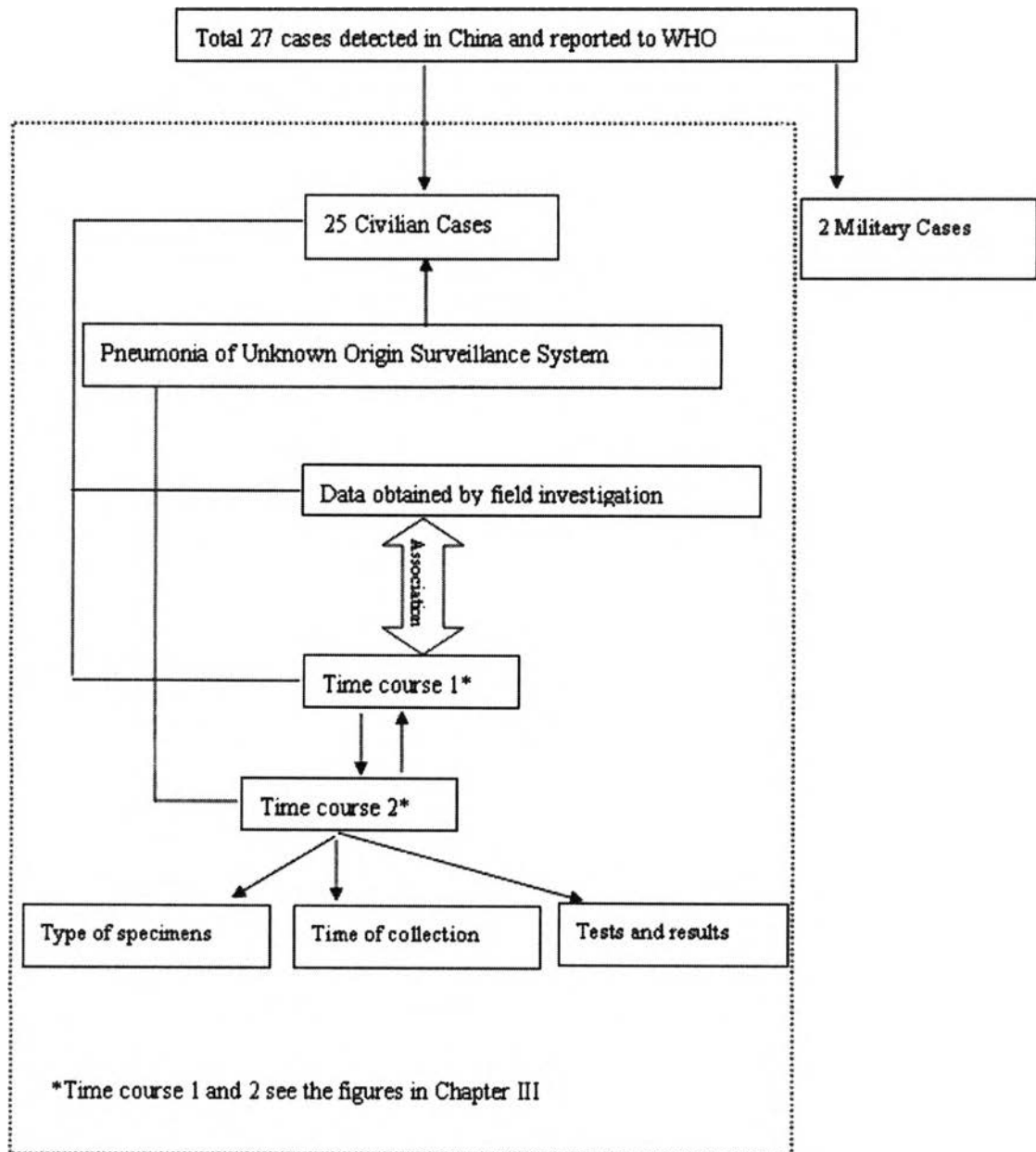

Figure 5: Conceptual framework of study design and data collection

H5N1 was confirmed by virus isolation, reverse transcriptase-polymerase chain reaction (RT-PCR) or microneutralization (MN) and hemagglutination-inhibition (HI) assay. 18 (73%) case patients resided in rural areas at the time of symptom onset and 10 cases that resided in urban areas. Fever and respiratory disease were present in all cases at hospital admission. Many case-patients had evidence of multi-organ impairment at admission, including hepatic, renal, and cardiac dysfunction. All case-patients had abnormal chest radiographs at admission.

To date, data are available on the clinical findings and clinical course of H5N1 disease since 1997 (Beigel et al., 2005), but not available on the time course of human with avian influenza A (H5N1) infections which has an important relationship with disease surveillance and case patient detection. Timely and thorough surveillance for avian influenza A (H5N1) in human beings is essential to monitor the disease situation and prevent possible person-to-person spread. By enabling prompt access to treatment, it may also improve the prognosis for H5N1 patients. Although country reports of laboratory-confirmed cases to WHO remain the official channel for reporting and international notification, these reports do not give sufficient information to allow the global community to assess the effectiveness of any individual country's surveillance system. Therefore, additional information is needed. Here we describe and analyze the time course of 27 civilian H5N1 cases in China, in order to provide such additional information, and to assist in increasing the timeliness and thoroughness of reporting of avian influenza

1.2 Research objectives

The objectives of this study are:

1. To describe time course with key indicators of human infection with avian influenza A (H5N1) in China between October 2005 and December 2007 by different dates of illness onset, first time seeking medical care, hospital's admissions, antivirus treatments, specimens collection, suspect case/case reporting, and outcomes of case patients.

2. To analyze time course and possible determinants of detection in human infection with avian influenza A (H5N1).

3. To compare sensitivities of different diagnostic laboratory tests, of tests conducted with different collected specimens (e.g., nasal swabs and sputum samples), and of the same test conducted at different times during follow-up.

1.3 Research questions

1. What are the indicators to identify the different durations of disease time course of human with avian influenza A (H5N1) in China? And what are the time intervals between different dates of illness onset, first time seeking medical care, hospital's admissions, antivirus treatments, isolation of suspected cases, specimen collection, case reporting, and outcomes in H5N1 patients?

2. What are the characteristics of time course distribution among humans with avian influenza A (H5N1) in China from 2005 to 2008? Are there any differences among the cases or are there existing factors impact on the disease time course?

3. What are the potential determinants (for example, demographic, level of education, structure of surveillance system) associated with cases reported in China? What are

the impact factors of public health surveillance system to reliably detect a H5N1 human case at the earliest possible stage?

4. What are the sensitivities of laboratory tests with various specimens among H5N1 case detection, and what is the sensitivity of same test on various specimens within the different time course of disease?

1.4 Research hypotheses

The time course of H5N1 detection in general population reflects the nature of evolution of disease as well as the capability of a surveillance system. Also, further understanding the sensitivities of different laboratory tests for various specimens will improve the conformation of H5N1 infection.

1. There are specific indicators assist to identify the different dates of disease time course of human with avian influenza A (H5N1) in China. Those indicators would benefit the future outbreak investigation in China as well as other countries.

2. The time intervals between different dates of illness onset, first time seeking medical care, hospital admissions, antivirus treatments, suspect case isolation, specimen's collection, case reporting, and outcomes of H5N1 patients are different.

3. The time course of avian influenza A (H5N1) detection, diagnosis, and initiation of treatment in China differs from that in other countries.

4. Mortality risk in avian influenza A (H5N1) may relate to the time course of detection, diagnosis, or initiation of treatment.

5. Factors potentially affecting case reporting, or diagnostic or treatment delay, include younger age group, males, patients from rural areas, lack of knowledge and having poultry outbreaks in the living area.

6. A higher yield Avian Influenza A (H5N1) Virus from clinical lower respiratory tract specimens from human infections with H5N1 in China and The earlier case is reporting would facilitate the specimens collection, thus, higher virus isolation will come as a sequence which will enhance the case confirmation and implement prevention strategy.

7. The surveillance system for avian influenza A (H5N1) human cases detection has been established in China, but the report of cases need to be enhanced by educate primary healthcare workers.

1.5 Variables to be studied

The following variables will be studied in this research:

1. Time course data with key indicators:

Days of first exposure, most recent exposure, type of exposure, days of illness onset and onset of symptoms; days of seeking medical care and diagnose or medicine; days of admission to hospitals and diagnoses, treatment or medicine in brief; days of transfer among hospitals and the level of each hospital admit to; days of specimen collection and type of specimens; days of laboratory test and results; days of suspect case reporting and report diagnoses; days of case confirmation and control measures; days of field investigation and control measures; Days of discharge or death.

2. Laboratory data

Specimens' type, date of collection, numbers of specimens, handling, transportation, test date, test method, results of tests.

3. Surveillance system structure and case reporting procedure

Frame work of surveillance system for H5N1 human case detection in China, purpose and objectives of the system, mean component of case reporting, population under surveillance, responsibilities of health care professionals

1.6 Definitions

Definition of Avian Influenza A (H5N1) Case Patients. A confirmed H5N1 case was defined as a patient with pneumonia or Influenza Like Illness (ILI) with laboratory evidence of H5N1 virus infection diagnosed by viral isolation, or reverse transcriptase-polymerase chain reaction (RT-PCR) test positive, or a 4-fold or greater increase in H5N1 antibody titer in paired acute and convalescent sera.(Atashili et al., 2006)

Definition of Pneumonia of Unknown Origin

A patient with all of the following criteria without specific laboratory diagnosis: (a) fever (temperature $\geq$ 38°C); (b) radiological evidence of pneumonia or acute respiratory distress syndrome (ARDS); (c) Normal white blood cell count (WBC) [range: 4 - 10 x 10^9/L) or leukopenia (WBC <4 x 10^9/L) or lymphopenia (lymphocyte count <0.8×10^9/L) at clinical presentation; and (d) absence of clinical improvement after treatment with broad spectrum antibiotics.

Definition of influenza-like illness (ILI)

A patient with fever $\geq$ 38°C and cough or sore throat, in the absence of any other confirmed diagnosis.

Definition of Incubation Period

The "incubation period" is defined as the time from infection to onset of clinical symptoms of disease