

H5N1/Highly Pathogenic Avian Influenza in Cambodia:

Evaluating poultry movement and the extent of interaction
between poultry and humans



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Submitted for the Degree of Doctor of Philosophy

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**University of London
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Abstract

Since 2003, highly pathogenic avian influenza (HPAI), subtype H5N1, has spread across the Asian, African and European continents at an exceptional rate. To date, H5N1 remains primarily a pandemic within poultry populations with limited onward transmission to humans. Since there have been a limited number of human cases throughout the world, epidemiologic uncertainties exist regarding the extent of contact necessary to result in successful transmission between infected poultry and humans. In this thesis I undertook two large-scale surveys to evaluate poultry movement and the extent of interaction between humans and poultry to better define the risks of sustained transmission of H5N1 in poultry and onward transmission to humans.

The thesis begins with a review of current knowledge on the epidemiology of HPAI, specifically subtype H5N1, and current options for its control worldwide and specifically within Cambodia. The first half of the thesis presents the methodology and results from a large-scale cross sectional survey of 3,600 rural subjects from 115 villages in six provinces throughout Cambodia. The results from this survey are used to explore animal ownership and husbandry, poultry mortality experienced and poultry mortality reporting, and the extent and frequency of poultry handling behaviours of subjects and how they differ by age and gender.

The second half of the thesis presents results from a second cross-sectional survey of 715 rural villagers, 123 rural, peri-urban and urban market sellers and 139 middlemen from six Provinces and Phnom Penh, which was conducted to evaluate poultry movement and trading practices. The results from this survey are used to construct poultry movement networks using social network analysis techniques, to identify critical points for surveillance and to understand the potential transmission and control of HPAI over this network and to identify a spatial model to predict poultry movements. Finally in the last chapter the key findings are presented and discussed in the context of HPAI transmission in the region.

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List of Appendices

Appendix A Published Manuscripts as the result of PhD work:

Ly S, **Van Kerkhove MD**, Hull D, Froehlich Y, Vong S. Human and Poultry Interaction, Rural Cambodia. *Emerg Infect Dis*, 2007;13(1)130-132.

Van Kerkhove MD, Ly S, Holl D, Guitian J, Mangtani P, Ghani A, Vong S. Frequency and types of contact with poultry and potential risk of H5N1 transmission to humans in rural Cambodia. *Influenza and Other Respiratory Viruses*, 2008; 2:155-163.

Appendix B Questionnaires for rural Cambodians:

Village chief, head of household, adult household members and children

Appendix C Interviewer Training Manual

Appendix D Consent Forms

Appendix E Ethics Approval Forms from LSHTM and the Cambodian Ministry of Health

Appendix F Questionnaires for poultry market sellers and middlemen

Abbreviations

AI	Avian Influenza
β	Transmission Risk Weighting Score
CDC	Communicable Disease Control Department, Ministry of Health
CFR	Case Fatality Rate
CP Company	Charoen Pokphand Poultry Company
DAHP	Department of Animal Health and Production
DIVA	Differentiating Infected from Vaccinated Animals
dpi	Days post inoculation
EIA	Enzyme Immunoassay
FAO	Food and Agriculture Organization of the United Nations
GPS	Global Positioning System
HA	Hemagglutinin
HI	Haemagglutination Inhibition
HPAI	Highly Pathogenic Avian Influenza
HPAI/H5N1	Highly Pathogenic Avian Influenza, Subtype H5N1
ILI	Influenza-Like Illness
IAV	Influenza A Viruses
IFA	Immunofluorescence Assay
IPC	Institute Pasteur of Cambodia
IQR	Interquartile Range
KAP	Knowledge, Attitudes and Practices
Kg	Kilograms
Kg Cham	Kampong Cham Province, Cambodia
Kg Speu	Kampong Speu Province, Cambodia
Km	Kilometre
LBM	Live bird markets
LPAI	Low Pathogenic Avian Influenza
LSHTM	London School of Hygiene and Tropical Medicine
MAFF	Ministry of Agriculture, Forestry and Fisheries, Cambodia
MN	Microneutralization
MoH	Ministry of Health
NA	Neuraminidase
NaVRI	National Veterinary Research Institute, Phnom Penh, Cambodia
OIE	World Organization for Animal Health
OR	Odds Ratio
PCA	Principal Components Analysis
PC1	Principal Component 1 (Practice Score 1)
PC2	Principal Component 2 (Practice Score 2)
PCR	Polymerase Chain Reaction
PPE	Personal Protective Equipment
PPS	Probability Proportion to (Population) Size Methodology
PV	Prey Veng Province, Cambodia
RNA	Ribonucleic Acid
RR	Rate Ratio
RT-PCR	Reverse transcriptase Polymerase Chain Reaction
SNA	Social Network Analysis
VAHW	Village Animal Health Workers
VN	Virus Neutralization
VSF	Vétérinaires Sans Frontières
UNICEF	United Nations Children's Fund
US	United States
US CDC	US Centers for Disease Control and Prevention
USHHS	US Health and Human Services
WHO	World Health Organization

Chapter 1 Introduction

1.1 Research Aims and Objectives of the PhD Thesis

The overall aim of this thesis is to evaluate poultry movement and the extent of interaction between humans and poultry in Cambodia to better define the risks of sustained transmission of H5N1 in poultry and onward potential transmission to humans.

H5N1 outbreaks in poultry populations have far outweighed the number of reported human cases of H5N1 (FAO 2008). Given that exposure to H5N1 infected poultry is believed to be the main route of transmission of H5N1 from poultry-to-humans (Writing Committee of the Second World Health Organization Consultation on Clinical Aspects of Human Infection with Avian Influenza 2008) and that a large proportion of the developing world may be living in close proximity to poultry (Epprecht & Robinson 2007; Gilbert et al. 2007), there is substantial risk for further human cases. To date, transmission of H5N1 from poultry-to-humans has been limited; however the extent of interaction between poultry and humans is unknown.

The first aim of my PhD thesis is to identify populations living in rural Cambodia with the highest H5N1 (or other subtypes of avian influenza) exposure potential.

Research Question 1: What is the frequency and extent of exposure to poultry in the general as well as occupationally exposed populations in Cambodia?

The specific objectives are to:

- Determine the extent and frequency of poultry handling behaviours of rural adult males, adult females and children and how they differ by age and gender;
- Determine the extent and frequency of poultry handling behaviours of poultry traders (i.e., poultry market sellers and middlemen); and
- Use risk assessment methods and the study subjects' patterns of contact with poultry to generate risk indices of potential H5N1 transmission to different populations in contact with poultry.

Additionally, despite their likely role in the circulation and spread of HPAI in South East Asia, little is understood about the poultry market chains, legal or illegal trade of poultry or the types and frequencies of contact that exist between rural people raising poultry, local markets and large-national poultry markets in the major cities. The connectedness of animal networks via poultry can lead to large and widespread epidemics of disease and an understanding of human and animal movement and their contact structures could be used to design more targeted surveillance activities and inform models of disease spread which could result in more cost-effective disease prevention and control (Dent et al. 2008; Green et al. 2008; Kiss et al. 2008; Truscott et al. 2007). Because trade of poultry may be responsible for some transmission of H5N1 within countries (Normile 2005a; WHO 2006-2009), controlling the movement of live poultry and poultry products could contain or reduce the spread of the virus.

The second aim of this thesis is to describe the current movements of poultry throughout Cambodia and determine how these movements influence the potential spread of HPAI at local, regional and national levels.

Research Question 2: How do current movements of poultry influence the potential spread of HPAI at local, regional and national levels? What are the implications of these movements for control and containment of H5N1 in poultry and/or human populations?

The specific objectives are to:

- Identify poultry selling markets and their trading characteristics in rural Cambodia and Phnom Penh;
- Identify market sellers and middlemen responsible for commercial trade of poultry;
- Characterize poultry trading practices of rural Cambodians, market sellers and middlemen;
- Identify and characterize the poultry (chickens and ducks) selling network in Southern Cambodia;
- Characterize the potential role of networks in HPAI/H5N1 virus circulation using social network analysis methods; and
- Identify a spatial model that can predict poultry movement patterns.

1.2 Structure of the PhD Thesis

Chapter 2 describes the current state of knowledge on the epidemiology of highly pathogenic avian influenza (HPAI), specifically subtype H5N1, with a focus on transmission in wild and domestic bird populations and the zoonotic transmission risk from poultry to humans. The chapter also reviews the importance of animal movement in disease circulation, current options for controlling H5N1 in poultry populations and in context of the PhD thesis presented, the chapter summarizes the occurrence of H5N1 in Cambodia prior to the initiation my field work in 2006. Chapter 3 presents the methodology of a large-scale cross sectional survey of 3,600 rural subjects from 115 villages in six provinces throughout Cambodia. Chapter 4 describes animal ownership and husbandry, poultry mortality experienced and reporting of rural Cambodians, as well as the study subjects understanding of avian influenza. The chapter also explores redefining FAO's poultry sectors in the context of countries with large sector 4 holdings and offers newly defined categories for such countries, which dominate some of Asia and most of Africa.

Chapter 5 describes the extent and frequency of poultry handling behaviours of subjects and how they differ by age and gender. Using risk assessment methods, patterns of contact with poultry were used to generate risk indices of potential H5N1 transmission to different populations in contact with poultry. Chapter 6 presents the results of a second cross-sectional survey of 715 rural villagers, 123 rural, peri-urban and urban market sellers and 139 middlemen from six Provinces and Phnom Penh, which was conducted to evaluate poultry movement and trading practices. This chapter describes the current movements of poultry throughout the study areas and examines how these movements influence the potential spread of HPAI at local, regional and national levels. In addition, the results of this study were used to inform the Cambodia's HPAI strategies. Chapter 7 explores the driving forces that may be behind poultry movement by using gravity model theory and Chapter 8 presents the key findings of the thesis, how the results of the thesis have been disseminated and used by local collaborators and interpretation of results in context of HPAI in the Mekong Delta Region.

1.3 Role of the Author

This PhD work has been developed and carried out by the doctoral candidate and principal investigator (PI), Maria Van Kerkhove, in collaboration with my PhD advisory committee, the Institut Pasteur du Cambodge (IPC; host institution), UNICEF (funding organization)

and the National Veterinary Research Institute (NaVRI), Ministry of Agriculture, Fisheries and Forestry (MAFF; collaborating institution).

All fieldwork for the studies was conducted in Cambodia between April 2006 and December 2007, under the field supervision of Sirenda Vong, MD, Head of Epidemiology Unit, IPC. The field work was led by me with assistance from IPC, Cambodian interviewers, village chiefs and district, provincial and national veterinarians.

All of the results presented in this thesis and manuscripts resulting from the PhD (Appendix A) were written under the supervision of Azra Ghani, Punam Mangtani, Javier Guitian and Sirenda Vong, but are entirely my own work. I received input from Azra Ghani, Tini Garske and James Truscott on the analysis using gravity model theory (Chapter 7).

Chapter 2 Highly Pathogenic Avian Influenza (HPAI/H5N1) and the Risk of Onward Transmission to Humans

Highly pathogenic avian influenza, subtype H5N1 (HPAI/H5N1) first crossed the species barrier in 1997 when an outbreak of 18 human cases resulting in six deaths was identified in Hong Kong (Claas et al. 1998; Lee et al. 1999). In late 2003, H5N1 crossed the species barrier a second time infecting a family from Hong Kong that had recently travelled to Fujian Province in China (WHO 2008b). Since 2003, H5N1 has been confirmed in domestic poultry and/or wild birds in 61 countries throughout Asia, Africa and Europe—largely in Vietnam, Thailand and Egypt (OIE 2008e)—and in 391 humans in 15 countries—largely in Indonesia and Vietnam (WHO 2006-2009).

The first half of this Chapter reviews the epidemiology of HPAI in poultry and humans, focusing on H5N1 but drawing on lessons learned from outbreaks of other highly pathogenic strains of avian influenza such as the H7 outbreaks in poultry and humans in the Netherlands and in poultry in Italy. The second half of this Chapter summarizes the situation of HPAI/H5N1 in Cambodia prior to the start of my PhD in early 2006 with some additional updates during the course of my studies.

2.1 Influenza A Biology

There are three types of influenza viruses – A, B and C – within the *Influenzavirus* genus and *Orthomyxoviridae* family. Only type A is capable of causing severe infections and pandemics in human populations (Webster et al. 1992), although type B can cause severe morbidity and mortality particularly in children (Jefferson et al. 2008). The central core of influenza A viruses contain eight single-stranded RNA gene segments surrounded by the surface glycoproteins hemagglutinin (HA) and neuraminidase (NA) (Figure 2-1) (de Jong et al. 2000; Lee et al. 2006; Oxford 2000). Influenza A viruses are classified into subtypes based on the antigenicity of HA and NA glycoproteins. There are 16 HA and nine NA subtypes. Only three HA (H1, H2, H3) and two NA subtypes (N1, N2) are known to have been widely present in humans (Horimoto & Kawaoka 2001).

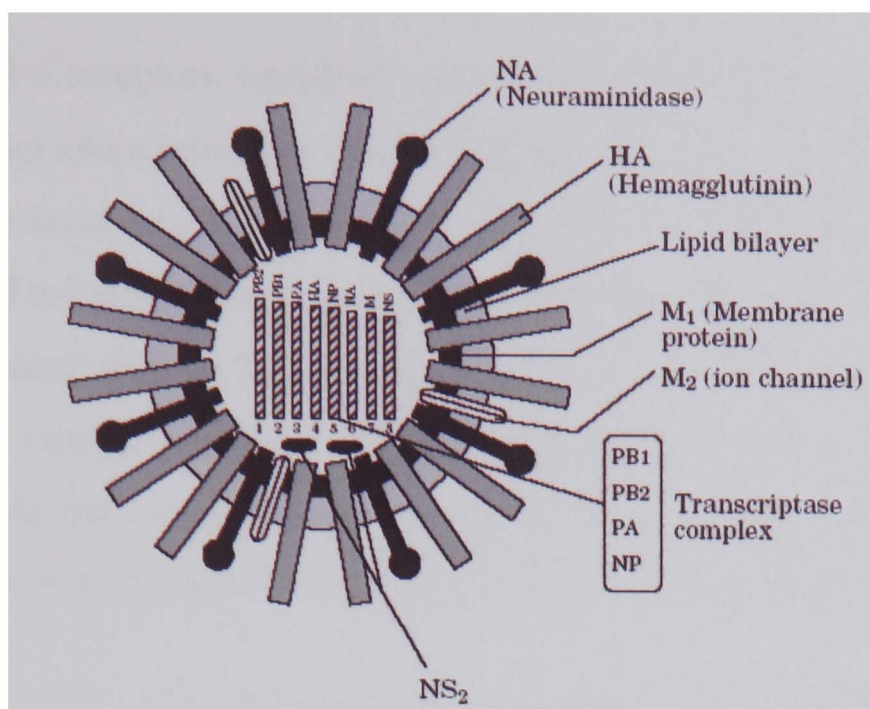


Figure 2-1 Illustration of the structure of influenza A virus

Source (de Jong et al. 2000)

Influenza A viruses can infect several animal species including birds, pigs, horses, seals, cattle, and whales (Table 2-1). The natural host of all HA and NA subtypes are aquatic birds mainly ducks, gulls and water birds (Horimoto & Kawaoka 2001; Webster et al. 1992; Webster et al. 2006a).

Table 2-1 Reservoir for HA and NA subtypes

Host	HA Subtypes	NA Subtypes
Human	H1, H2, H3, H5, H7,	N1, N2, N3, N7
Pig	H1, H3, H4, H9	N1, N2
Waterfowl	All 16 subtypes	All 9 subtypes
Horse	H3, H7	N7, N8
Seal	H4, H7	N7
Cattle	H3	N2
Whale	H3, H13	N2, N9
Cat, Tiger	H5	N1

The variability of influenza A viruses depends on the evolution of the virus through point mutations (antigenic drift) and genetic reassortment (antigenic shift) (Alexander & Brown 2000; Horimoto & Kawaoka 2001). Minor changes in the surface glycoproteins occur from point mutations due to the absence of proofreading mechanisms of RNA molecules as the virus replicates in the host. These point mutations occur often resulting in annual variation in the human influenza strains circulating the globe. It is these changes that require the production of new human seasonal influenza vaccines each year (Jennings & Read 2006).

Humans are naturally protected from avian influenza viruses because we lack certain receptor binding sites (α 2-3 receptors) in our respiratory tracks that are required for infection to occur.

Humans possess α 2-6 receptors, which are susceptible to human influenza viruses (e.g., H1N1, H3N2) but not avian influenza viruses. Pigs are susceptible to both human and avian influenza viruses because they possess receptors for both avian and human influenza viruses (α 2-3 receptors and α 2-6 receptors, respectively), and therefore can serve as an intermediate host (i.e., mixing vessel) (Figure 2-2). Antigenic shift results from the reassortment of two distinct influenza A viruses (e.g., avian and human influenza viruses) within a single host (e.g., pigs) and represents a major change in viral composition. This can result in the formation of novel viruses (Capua & Alexander 2002; Horimoto & Kawaoka 2001; Tambyah & Leung 2006).

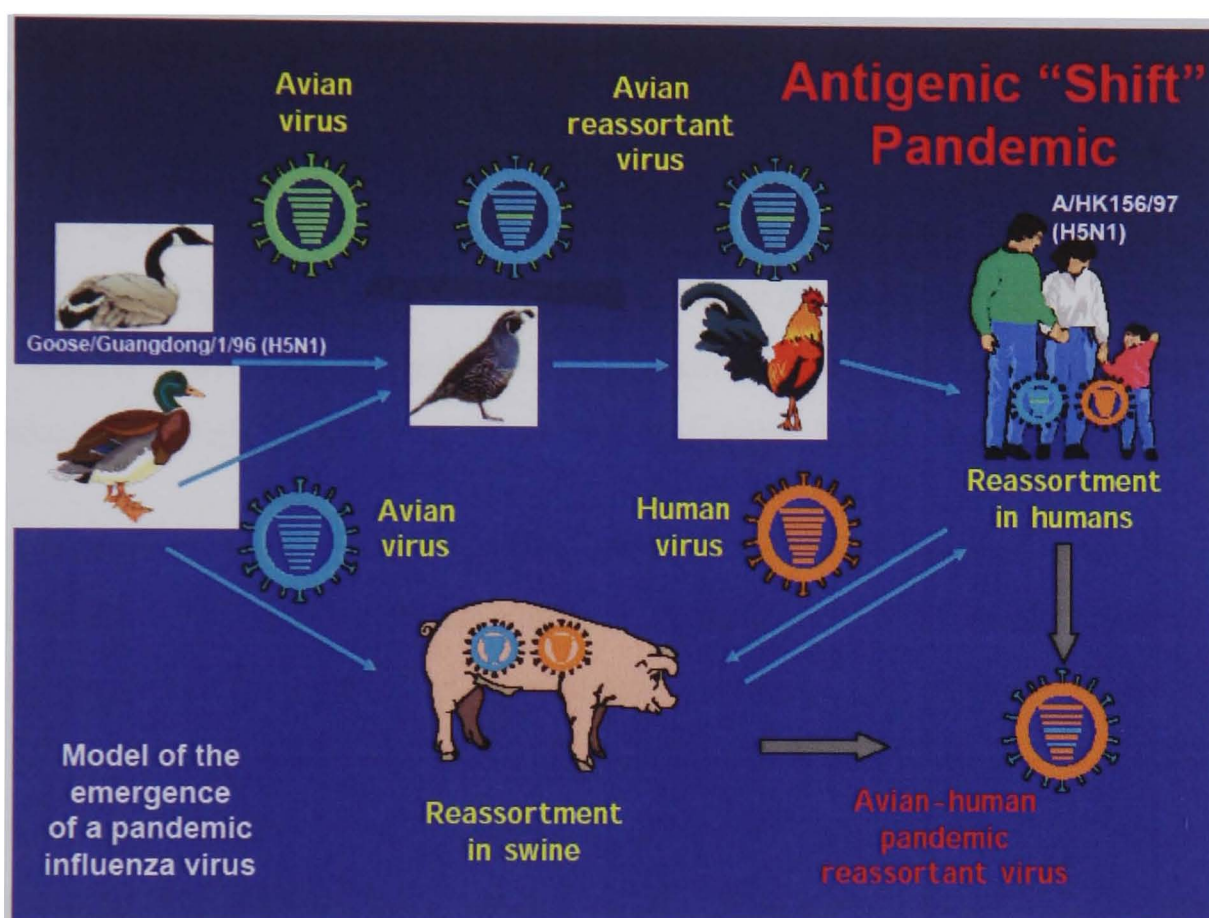


Figure 2-2 Illustration of antigenic shift or genetic reassortment of influenza A viruses

Source: Centers for Disease Control; image courtesy of S. Vong, IPC

2.1.1 Clinical Manifestations of H5N1 in Birds

Influenza A viruses occurring in birds are collectively termed avian influenza. Avian influenza strains are categorized as having high (HPAI) or low pathogenicity (LPAI) based on the severity of disease and mortality caused in birds. LPAI strains are capable of mutating into HPAI as occurred in the Italian H7N1 outbreak in 1999-2000 (Capua & Alexander 2006; Capua & Marangon 2000; Mannelli et al. 2006).

HPAI/H5N1 has been further categorized into genetic clades. Phylogenetic analysis of the H5 NA genes circulating since 2003 indicate that Clade 1 strains have been circulating in Thailand, Vietnam and Cambodia whereas Clade 2 (and several subclades 2.1-2.3) have been circulating in Indonesia (subclade 2.1), Europe, the Middle East and Africa (subclade 2.2) and China, Japan and South Korea (subclade 2.3) (WHO 2005).

HPAI strains (always of the H5 or H7 subtypes) replicate rapidly in the gastrointestinal tract of birds and can systematically spread and replicate in multiple organs often resulting in rapid death (Capua & Alexander 2006; Mannelli et al. 2006). Chickens (order *Galliformes*) are more susceptible to influenza A viruses than ducks, geese and swans (order *Anseriformes*) and therefore are more likely to be diseased and die from infection (Swayne & Suarez 2000).

Symptoms of HPAI/H5N1 in birds range from asymptomatic, mild disease (anorexia, depression, weight loss) to severe neurological symptoms (e.g., tremors, shaking, lack of coordination, spinning, seizures) and sudden death (Pantin-Jackwood et al. 2007). Severe disease is usually caused by systemic virus replication affecting organs and tissues (Ellis et al. 2004a; Hulse-Post et al. 2005; Sturm-Ramirez et al. 2004; Sturm-Ramirez et al. 2005).

Experimental studies, which typically infect animals with high doses of virus, have demonstrated that chickens are almost always susceptible to HPAI/H5N1 infection with 80-100% mortality occurring within 1-5 days post inoculation (dpi) (Saito et al. 2009; Spickler et al. 2008; Tian et al. 2005; Webster et al. 2006b). Experimental evidence has shown that the pathogenicity and mortality of HPAI/H5N1 in ducks has changed since 2002 and varies depending on the infecting strain (Chen et al. 2004; Hulse-Post et al. 2005; Pantin-Jackwood et al. 2007; Sturm-Ramirez et al. 2005).

Mortality can occur faster in chickens (within 1-5 days) (Tian et al. 2005; Webster et al. 2006b) than ducks (6-7 days) (Beato et al. 2007; Hulse-Post et al. 2005; Tian et al. 2005). Morbidity and mortality of HPAI/H5N1 infection in ducks also varies by age (Pantin-Jackwood et al. 2007). During an outbreak of HPAI (H5N1) commercial domestic ducks in South Korea in 2003-2004, morbidity and mortality was higher in younger ducks as compared to older animals (Kwon et al. 2005).

Clinical signs are almost always present in chickens infected with HPAI/H5N1 with onset typically from 2-5 dpi until death (Mase et al. 2005; Shortridge et al. 1998; Tian et al. 2005; Tumpey et al. 2002). Tracheal viral shedding and cloacal/faecal viral shedding have been

experimentally shown to begin on or before day 2 (1-3) dpi (Bublout et al. 2007; Perkins & Swayne 2001; Swayne & Beck 2005; Tian et al. 2005).

Although the susceptibility of chickens to HPAI/H5N1 almost always leads to clinical symptoms and death, the susceptibility of wild birds and domestic ducks depends on several factors including the circulating strain (Hulse-Post et al. 2005; Sturm-Ramirez et al. 2005) and the age of the ducks. This indicates that the pathogenicity with HPAI/H5N1 in ducks is somewhat inconsistent (Pantin-Jackwood et al. 2007) and may be a factor in the observed differences in geographic distribution of poultry outbreaks.

In experimental studies of ducks, the onset of clinical symptoms occurs 2-10 dpi (Beato et al. 2007; Middleton et al. 2007) and oropharyngeal and cloacal shedding can occur from 2-7 or up to 11-17 dpi (Hulse-Post et al. 2005; Shortridge et al. 1998). The infectious period of ducks is estimated to be 4.3 days (95% CI 3.8-4.8) (van der Goot et al. 2008). Virus titers have been found to be highest 1-3 dpi and reduce to undetectable levels by 13-20 dpi (Hulse-Post et al. 2005; van der Goot et al. 2008). Typically virus shedding is higher in symptomatic ducks. In experimental and in field settings, H5N1 virus has been detected in cloacal, tracheal and blood samples of asymptomatic ducks (Vong et al. 2006).

In wild ducks and waterfowl, H5N1 has been found to replicate in the gastrointestinal tract and can shed the virus for up to 30 days (Claas et al. 1998; Sturm-Ramirez et al. 2005). Some avian viruses are shed in higher doses in the pharynx than in faeces of wild ducks and mallards (Keawcharoen et al. 2008; Normile 2006; Sturm-Ramirez et al. 2005). However, many LPAI are shed at higher titres in faeces.

The stability of HPAI/H5N1 in poultry faeces is not well understood. Experimental evidence suggests that H5N1 loses infectivity in chicken faecal manure within 24 hours at 25°C and within 15 minutes at 40°C (Chumpolbanchorn et al. 2006), indicating that the infectiousness of contaminated faecal manure may be shorter in warmer climates. However, another study suggests that H5N1 is viable in faeces for 2 days at 37°C (Shortridge et al. 1998) highlighting that further experimental study is necessary to understand the persistence of H5N1 in the environment under various environmental conditions.

Data on the persistence of HPAI/H5N1 virus in tissues is limited. An experimental study of ducks challenged with HPAI/H5N1 demonstrated that the virus is detectable in breast and thigh tissue at 3-7 dpi, in the liver and intestine at 3-4 dpi and in the lung at 3-6 dpi. An

experimental study of chickens challenged with HPAI/H5N1 found virus detectable in the trachea, lung, bone, breast meat and thigh tissue at 1-5 dpi (Swayne & Beck 2005). These results suggest that systemic infection occurs at a faster rate in chickens than ducks and provides insight on why HPAI appears to be more virulent in chickens.

Since wild ducks, domestic ducks and geese infected with HPAI/H5N1 can be asymptomatic, they may act as silent vectors for transmission and represent a major challenge in controlling the spread of HPAI (Chen et al. 2004; Hulse-Post et al. 2005; Keawcharoen et al. 2008).

2.1.2 Clinical Manifestations of HPAI in Humans

The pathogenicity of HPAI/H5N1 and HPAI/H7N7 in humans ranges from undetected asymptomatic or sub-clinical to severe disease resulting in death. Although the CFR of HPAI/H5N1 is high, this may be an overestimate of the true CFR since relatively few seroprevalence studies have been carried out to determine the number of subclinical or asymptomatic cases in countries affected by H5N1 outbreaks in humans, domestic or wild poultry populations.

The incubation period of H5N1 in humans is believed to be less than 7 days (range: 2-9 days) (Areechokchai et al. 2006; Huai et al. 2008; Writing Committee of the Second World Health Organization Consultation on Clinical Aspects of Human Infection with Avian Influenza 2008). The first symptoms of H5N1 disease—typical of seasonal influenza (fever, dyspnoea, cough, sore throat) and pneumonia but sometimes including gastrointestinal symptoms (abdominal pain, diarrhoea, or vomiting)—usually appear within 1-4 days after exposure, although they can take up to 8 days to appear. Among severely affected patients, severe respiratory distress syndrome can occur as well as bilateral pneumonia and multiorgan failure (Gambotto et al. 2008; Uyeki 2008; Writing Committee of the Second World Health Organization Consultation on Clinical Aspects of Human Infection with Avian Influenza 2008).

The pathogenicity of HPAI/H7N7 in humans following an outbreak in commercial poultry farms in the Netherlands resulted in 89 infected subjects who suffered mostly from mild illness including conjunctivitis (87.6% n=78), influenza like illness (2.2% n=2), or both conjunctivitis and influenza like illness (5.6% n=5). However one subject (1.1%) died of acute respiratory distress syndrome and pneumonia (Fouchier et al. 2004).

2.1.3 H5N1 Detection Methods

H5N1 infection can be detected through virologic and/or serologic testing methods. Serological tests (e.g., haemagglutination inhibition [HI] test, microneutralisation test, agar gel diffusion [AGID] test, enzyme-linked immunosorbent assay [ELISA]) detect antibodies indicating that an individual or bird has been infected in the past but cannot determine when infection occurred and are therefore indirect markers for infection (Katz et al. 1999; Rowe et al. 1999; Suarez & Schultz-Cherry 2000). Virological testing (e.g., rapid antigen detection tests, polymerase chain reaction [PCR] for nucleic acid detection, virus isolation after inoculation into cell cultures or embryonated eggs) assesses the presence of influenza A viruses and allows subsequent identification of specific viral subtypes (Chen et al. 2007).

Typically, suspect specimens are first tested to determine the presence of influenza A viruses or influenza A antibodies. If positive for influenza A virus or M gene detection, specimens undergo further testing to determine the subtype of the infecting strain (e.g., H5N1, H9N2, H3N2, etc). There are various tests that can be used to identify the presence of H5N1 virus. However, some methods are not appropriate for all settings because most techniques require highly trained staff to carry out the tests, and others also require bio-safety level 3 laboratories (BSL-3) because they involve handling live H5N1 viruses (e.g., virus isolation, microneutralisation tests) (Peiris et al. 2007).

2.1.3.1 Sample collection

From all suspected H5N1 human cases, guidelines from WHO recommend collecting samples from the upper respiratory tract (e.g., nasopharyngeal and/or throat swabs) and blood samples (for serology and/or nucleic acid detection). If the patient is hospitalized and intubated, samples from the lower respiratory tract (e.g. tracheal aspirates, broncho-alveolar lavage) should be collected (WHO 2006f). For suspected H5N1 in poultry populations, guidelines from OIE recommend collecting oropharyngeal samples and cloacal samples (or fresh faeces) from live birds, and organ tissue (e.g., trachea, lungs, air sacs, intestine, spleen, kidney, brain, liver and heart) from dead birds (Alexander 2008).

Throat or nasopharyngeal swabs from suspect humans and oropharyngeal or cloacal samples from suspect birds should ideally be taken as soon as possible for the detection of H5N1 virus (Alexander 2008; WHO 2006f). Because antibodies require a few days to a week to develop in birds (Suarez & Schultz-Cherry 2000) and sometimes more than 14 days to develop in

humans (Katz et al. 1999; Rowe et al. 1999), the timing of serum sample collection for anti-H5N1 antibody detection should be considered as antibodies are not immediately present following infection.

2.1.3.2 Detection of influenza A viruses and anti-H5N1 antibodies

Detection in suspect cases: The following procedures can be used to detect H5N1 virus from human and poultry specimens.

Rapid antigen tests are useful tools for influenza A virus screening. For humans specimens, rapid antigen detection using immunofluorescence assay (IFA) or enzyme immunoassay (EIA) methods are used to detect exposure to influenza A viruses (Peiris et al. 2007). AGID tests and ELISA are used to test for exposure to influenza A viruses from poultry specimens (Suarez & Schultz-Cherry 2000). These tests cannot distinguish between subtypes, rather they detect past or current exposure to all subtypes of avian and human influenza A viruses (Chan et al. 2007; Xu et al. 2005).

Several rapid influenza A antigen detection tests are available for field investigations of poultry outbreaks. An evaluation of five commercially available influenza A and H5 specific rapid antigen detection tests used during poultry outbreak investigations in Hong Kong between 2001 and 2003 revealed that the sensitivity of the detection tests (i.e., the proportion of true positives identified correctly) were higher when used on diseased bird specimens than for specimens from birds that appeared healthy. These results demonstrate that rapid antigen detection tests may be more appropriate for quick detection of H5N1 during outbreaks rather than routine surveillance of poultry flocks (Chua et al. 2007).

Furthermore, the clinical sensitivity and specificity of rapid antigen tests vary depending on the species tested (Chen et al. 2008; Peiris et al. 2007) and are poor for the detection of H5N1 in human and animal specimens (Beigel et al. 2005; Kandun et al. 2006; Oner et al. 2006). Therefore further testing is required to confirm the result and to determine the subtype of the virus present in human or poultry specimens.

RT-PCR or virus isolation in chicken embryos are methods used to test for the presence of H5N1 (Alexander 2008; Chan et al. 2007; WHO 2007). Viruses can be cultured using egg inoculation methods in which the allantoic fluid of embryonated fowl eggs are inoculated with specimens and incubated at 35-37°C for several days. Virus culture is typically

considered the gold standard for the detection of H5N1, but these methods are labour intensive and require BSL-3 laboratories (Peiris et al. 2007). RT-PCR can detect viral RNA and can specifically test for the presence of the H5N1 subtype by using specific primers targeting H5 and N1 genes within a few hours, thus this method is used most often to test for the presence of H5N1 (Chen et al. 2007; Lee et al. 2001). The World Health Organization only recognizes positive RT-PCR test results for human samples from WHO Collaborative Centres for Influenza, WHO H5 Reference Laboratories and from some National Influenza Centres (WHO 2008a).

Population Screening: Because immunity to influenza is long-lived, the presence of anti-H5N1 antibodies can be used to indicate that an individual or animal has previously been infected with an influenza virus and thus are useful for population screening. Serological tests for the presence of influenza A antibodies in human specimens include the HI, enzyme immunoassay (EIA), microneutralisation and virus neutralization tests (Katz et al. 1999; Rowe et al. 1999). HI, neuraminidase inhibition (NI) tests, AGID and ELISAs are methods used for the detection of influenza A antibodies in poultry (Shafer et al. 1998; Suarez & Schultz-Cherry 2000; Swayne et al. 1998; Zhou et al. 1998).

Human sera tested using an H5N1 virus specific microneutralisation assays are considered positive for anti-H5N1 neutralizing antibodies when titers are $\geq 1:80$ (Katz et al. 1999). For poultry specimens, sera is considered positive for anti-H5N1 antibodies when titers are $\geq 1:16$ (OIE 2005b). Human sera that test positive for anti-H5N1 antibodies are then tested using Western Blot techniques or HI tests using horse red blood cells. Sensitivity and specificity is highest when a combination of microneutralisation and Western Blot testing techniques are used (sensitivity 80-88%, specificity 96-100% depending on the age of the patient) (Rowe et al. 1999). The WHO requires a positive test result for both microneutralisation and confirmation with Western Blot or HI to be considered positive for anti-H5 antibodies (Katz et al. 1999; Rowe et al. 1999; WHO 2007).

2.2 Epidemiology and Transmission of HPAI/H5N1 in Birds

2.2.1 History of HPAI Pandemics in Birds

All strains of influenza A viruses naturally infect a large variety of wild birds, including wild ducks and waterfowl, but do not usually cause disease (Horimoto & Kawaoka 2001).

However, there have been several instances of major outbreaks of HPAI in poultry over the

last two and a half decades (Table 2-2) (CIRDAP 2006; Horimoto & Kawaoka 2001). HPAI/H5N1 was first detected in Hong Kong in 1997, but since 2003, HPAI/H5N1 has been confirmed in birds in 61 countries in Asia, Africa and Europe (Figure 2-3) (FAO 2008).

Table 2-2 Major outbreaks of HPAI (H5, H7) in poultry

Year	Location	Subtype	Approximate number of poultry culled or dead
1983	PA, USA	H5N2	17 million (culled)
1994-2003	Mexico	H5N2	1 billion
1995-2003	Pakistan	H7N3	3.2 million (dead)
1997	Hong Kong	H5N1	1.5 million (culled in 3 days)
1999-2000	Italy	H7N1	16 million birds (culled)
2003	The Netherlands	H7N7	30 million (killed)
2004	British Columbia, Canada	H7N3	>19 million (culled)
2003-present	Asia, Europe, Africa	H5N1	220+ million (culled or dead)

Source: (CIRDAP 2006; Horimoto & Kawaoka 2001)

2.2.2 Expanding Geographic and Host Range of H5N1

Since 2003, the geographic and host range of HPAI/H5N1 has spread. Figure 2-3 illustrates the countries which have reported H5N1 outbreaks in wild and domestic bird populations since 2003.

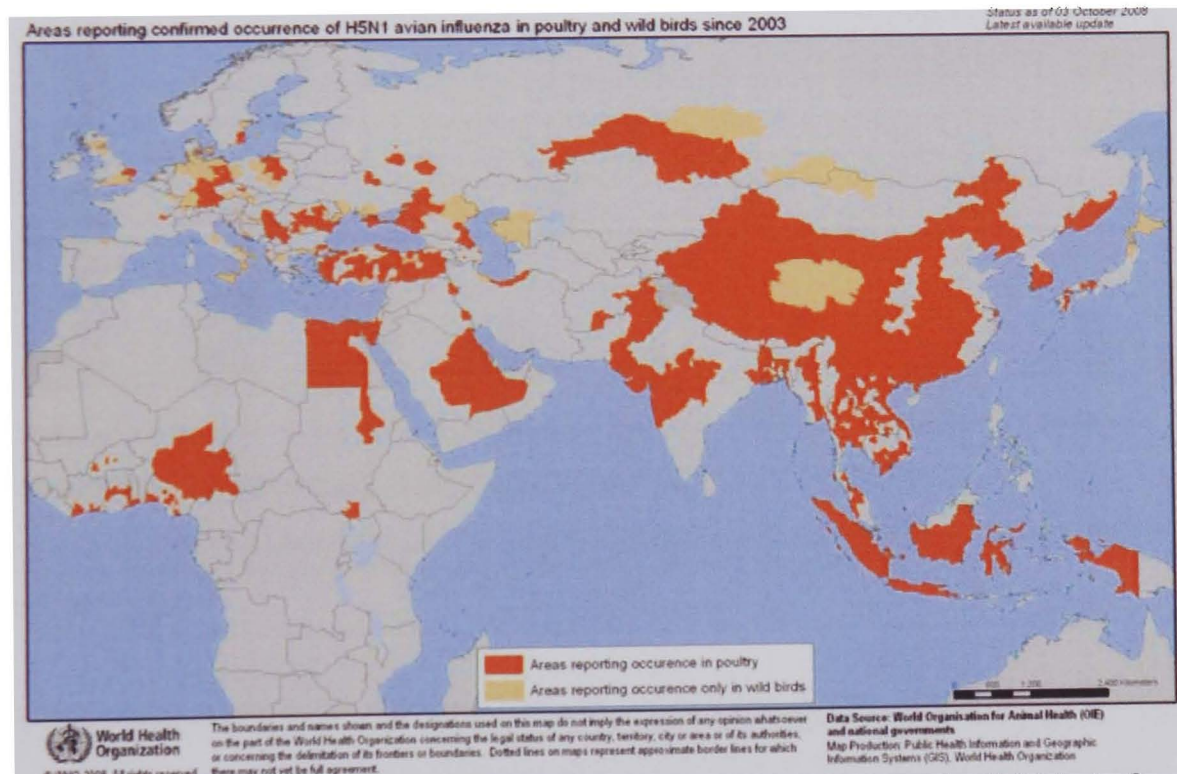


Figure 2-3 Countries reporting confirmed H5N1 in (left) domestic and wild birds from 2003 to 3 October 2008 and (right) humans from 2003 to 10 Sept 2008

Source: (WHO 2006-2009)

Approximately 6,500 H5N1 poultry outbreaks have been reported thus far, resulting in hundreds of millions of poultry culled (FAO 2008; OIE 2009a). Most outbreaks have been reported in Asia (65.6% of the outbreaks reported), and to a lesser extent in Africa, the

Middle East and Europe (FAO 2008). No outbreaks of H5N1 in domestic or wild birds have been reported in Australia, the Pacific Islands or the Americas.

The numbers of reported outbreaks reported in 2003-2008 among countries with the ten highest numbers of total outbreaks according to the World Organization for Animal Health (OIE) are shown in Figure 2-4. This graph also shows the cumulative number of reported outbreaks according to OIE (turquoise bar) and Food and Agriculture Organization (FAO) (orange bar), which vary significantly from each other making it difficult to fully understand the extent of outbreaks in wild and domestic bird populations. Differences in rates of detection of HPAI/H5N1 between countries may depend on the active and passive HPAI surveillance systems established and whether the focus of the surveillance system in place, if any, is on the commercial or backyard sector of poultry production. It has been suggested that it is more likely that HPAI will be detected in commercial farms as opposed to backyard flocks (Graham et al. 2008).

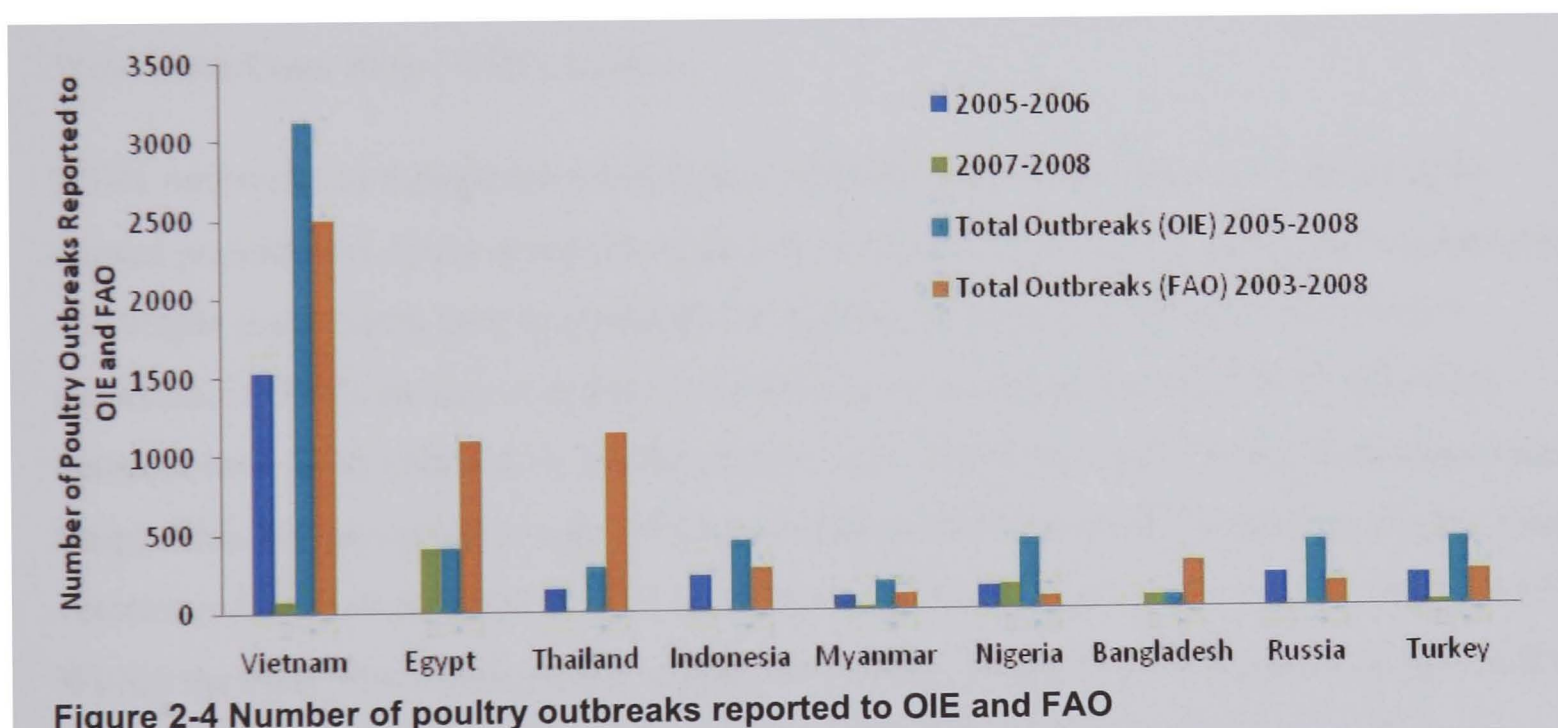


Figure 2-4 Number of poultry outbreaks reported to OIE and FAO

Note: Only countries that have the ten highest numbers of cumulative outbreaks according to OIE are included in this figure. Also note the higher number of reported outbreaks reported by OIE despite a smaller number of years included in the total
 Source: (FAO 2008; OIE 2009a)

HPAI/H5N1 was first detected in a goose in Guangdong Province in China in 1996 and spread to Hong Kong in 1997. In late 2003, H5N1 was first detected in a family from Hong Kong that had recently travelled to Fujian Province in China. Within the first six months of 2004, H5N1 was reported among poultry in Korea, Thailand, Vietnam, Cambodia, Laos, Japan, and Indonesia. Between July 2004 and July 2005, H5N1 was repeatedly detected in poultry in Thailand, Hong Kong, Indonesia, Vietnam and Cambodia (WHO 2008b). During this same time period, H5N1 expanded its host range to dogs, palm civits, ferrets, mice, and

small and large cats (Webster et al. 2007). Natural infection of HPAI/H5N1 was identified in tigers in a Thailand zoo who were likely infected from being fed contaminated poultry (Thanawongnuwech et al. 2005; WHO 2008b).

Since 2003, widespread outbreaks in domestic ducks in China may have led to the endemic situation in ducks in many countries throughout South East Asia (Chen et al. 2004; Hulse-Post et al. 2005). Additionally, human cases were often identified before outbreaks in poultry within many countries in Asia. This delayed detection may have also contributed to the endemic or recurrent situation in these countries (Sims 2007).

HPAI/H5N1 was first detected in Europe in July 2005 in Russia and in the Middle East in early 2006. Within eight months (July 2005- February 2006), H5N1 spread to domestic or wild poultry in 22 countries/territories including Kazakhstan, Turkey, Mongolia, Romania, Ukraine, the United Kingdom, Iraq, Italy, Slovenia, Kuwait, Bulgaria Croatia, Egypt, France, Germany, Austria, Hungary, Bosnia-Herzegovina, Slovakia, Azerbaijan, Georgia, and the West Bank/Gaza Strip (WHO 2008b).

H5N1 outbreaks in Europe have been more sporadic and to date, have only occurred in animal populations. Early detection in these countries is likely due to sufficient infrastructure and ample preparation time to establish surveillance systems for the early detection of incursion of H5N1 in their countries. Conversely, some countries where H5N1 has been detected have been affected by conflict or war (e.g., Afghanistan, Pakistan, West Bank/Gaza Strip). This has prevented proper HPAI surveillance due to limited financial resources, weak veterinary infrastructure and lack of access to some areas within these countries (Sims 2007). Within the Near East/North Africa region, the greatest number of outbreaks have occurred in Egypt, which has had outbreaks confirmed in poultry populations from almost all administrative regions in the country (MOH 2007).

In sub-Saharan Africa HPAI/H5N1 was first detected in Nigeria (Joannis et al. 2006)—possibly transmitted to the country through migratory birds or trade of live day-old chickens (Cecchi et al. 2008; Ducatez et al. 2006)—in January 2006 and has sporadically spread to domestic and/or wild birds in Cameroon, Burkina Faso, Sudan, Cote d'Ivoire, Djibouti, and Benin (WHO 2008b). Only two human cases of H5N1 have been identified throughout the whole of Africa, which occurred in Nigeria in early 2007 and in Djibouti in 2006. Since 2007, no further outbreaks in poultry and/or humans have been reported in Nigeria and no human

cases have been reported from any of the above named countries that have reported H5N1 outbreaks in poultry populations.

2.2.2.1 A Role for Wild Birds in Geographic Spread?

In 2005, H5N1 outbreaks occurred in wild migratory birds in Qinghai Lake, China (Chen et al. 2005). Because wild birds are believed to be the main reservoir of H5N1 and since all infected birds excrete high concentrations of virus in faeces (Claas et al. 1998; de Jong et al. 2000; Sturm-Ramirez et al. 2005; Webster 2002), it has been suggested that migratory birds are responsible for transporting HPAI/H5N1 to domesticated poultry in some countries in Asia (e.g., Japan, Republic of Korea (Sims 2007)) and to many countries in Europe, the Middle East and Africa (Figure 2-5) (Chen et al. 2005; Normile 2005a).

This suggestion is highly contested since data on wild bird outbreaks is largely incomplete and often incorrect (Yasue et al. 2006). It also assumes that infected birds are asymptomatic during migration since the impact of the disease on the fitness of the bird would be significant (Webber & Stillanakis 2007). Thus, it is unlikely that infected and symptomatic or asymptomatic, infected and virus-shedding birds are physically capable of carrying the virus over long distances (Webber & Stillanakis 2007).

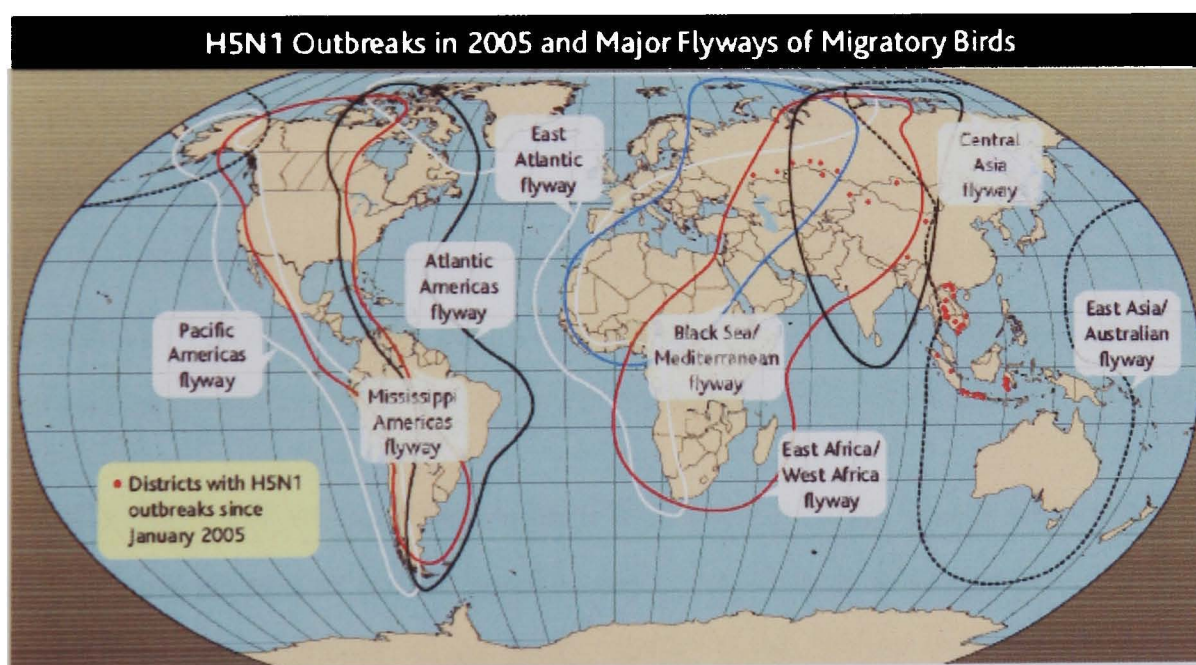


Figure 2-5 Major pathways of migratory birds

Source: (Normile 2005a)

Studies of wild birds and HPAI have largely focused on large scale influenza A (LPAI and HPAI) surveillance programs and have been implemented in various parts of the world including China, Northern Europe, North America, and Africa (Gaidet et al. 2007; Krauss et al. 2007; Parmley et al. 2008). Surveillance activities attempt to understand the viral-host

ecology, geographic dispersion (spatial patterns Figure 2-5), seasonality (temporal patterns) and host range of influenza A virus strains (Munster et al. 2007). Studies have included the sampling of tens of thousands of wild birds and have identified several previously unknown hosts for influenza A viruses (Munster et al. 2007) indicating that the natural host range of influenza A viruses has been expanding. Surveillance studies have suggested that wild migratory birds may have been responsible for the introduction of HPAI/H5N1 into western Europe (Bragstad et al. 2007; Starick et al. 2008) and Africa (Cecchi et al. 2008), demonstrating that wild birds may have had a role transmitting H5N1 between continents.

2.2.4 Animal-to-Animal Transmission of H5N1

Animal-to-animal transmission of H5N1 can be direct via the faecal-oral route (OIE 2008a) or indirect through contaminated feed, clothing, and equipment (fomites) (FAO 2005). Live markets may also be an important reservoir for H5N1 (Woo et al. 2006), as seen in H5N1 outbreaks in Vietnam, Thailand and Hong Kong (Amonsin et al. 2008; Kung et al. 2003a; Kung et al. 2007; Nguyen et al. 2005; Wang et al. 2006) (discussed later).

Movements of domestic poultry may also play a substantial role in viral spread. A study of the spatial distribution of HPAI outbreaks in Thailand showed a strong relationship between free-grazing ducks in rice fields and viral spread (Gilbert et al. 2006). Large bodies of water such as lakes that serve as resting places for wild aquatic birds may also play a role in transmission (Horimoto & Kawaoka 2001) because all birds shed virus in faeces (de Jong et al. 2000; Sturm-Ramirez et al. 2005; Webster 2002). Experimental evidence has suggested that influenza A viruses are detectable in water and wet faeces for up to 6 days at 37°C (Brown et al. 2007b) and H5N1 can survive in carcasses for several days at room temperature and longer in cooler (+4°C) temperatures (OIE 2008a; WHO 2006-2009).

It is also possible that trade of commercial and domestic poultry and poultry products, often occurring across long distances is responsible for transmission between and within countries (Chen et al. 2005; Normile 2005a; Sims 2007; WHO 2006-2009). Transmission is also likely to be occurring between wild and domestic bird populations in both directions (Normile 2006).

2.2.5 Current Options for Controlling H5N1 in Poultry Populations

Control methods for HPAI/H5N1 in poultry focus on reducing between-flock and poultry-to-human transmission and include increasing biosecurity measures, restricting poultry

movement, culling or stamping out infected and/or susceptible flocks, and prophylactic or emergency vaccination of at-risk poultry.

Before discussing each of these control options, it is important to describe how FAO categorizes the poultry production systems. FAO has classified poultry production into four sectors (Table 2-3) in which Sector 1 is described as the poultry production system for “industrial integrated” production system; Sectors 2 and 3 describe “commercial poultry production system” with decreasing levels of biosecurity, respectively; and Sector 4 describes “village or backyard poultry production” (FAO 2006).

Table 2-3 FAO defined poultry production sectors

Poultry Production System	Industrial and integrated production	Commercial poultry production		Village or backyard production
Sectors	Sector 1	Sector 2	Sector 3	Sector 4
Biosecurity	High	Mod-High	Low	Low
Market outputs	Export and urban	Urban/rural	Live urban/rural	Rural/urban
Dependence on market for inputs	High	High	High	Low
Dependence on goods roads	High	High	High	Low
Location	Near capital and major cities	Near capital and major cities	Smaller towns and rural areas	Everywhere. Dominates in remote areas
Birds kept	Indoors	Indoors	Indoors/Part-time outdoors	Out most of the day
Shed	Closed	Closed	Closed/Open	Open
Contact with other chicken	None	None	Yes	Yes
Contact with ducks	None	None	Yes	Yes
Contact with other domestic birds	None	None	Yes	Yes
Contact with wildlife	None	None	Yes	Yes
Veterinary service	Own Veterinarian	Pays for veterinary service	Pays for veterinary service	Irregular, depends on govt vet service
Source of medicine and vaccine	Market	Market	Market	Government and market
Source of technical information	Company and associates	Sellers of inputs	Sellers of inputs	Government extension service
Breed of poultry	Commercial	Commercial	Commercial	Native
Food security of owner	High	Ok	Ok	From ok to bad

Adapted from (FAO 2006): The values describing each criteria have not been modified.

HPAI/H5N1 has been detected primarily among Sector 4 poultry holdings in Asia and Africa (e.g., (Gilbert et al. 2006; OIE 2006b; Tiensin et al. 2007; Vong et al. 2006), but has affected commercial poultry farms in many countries (De Benedictis et al. 2007; Kwon et al. 2005; Mase et al. 2005; Team 2007a; Team 2007b). The options for controlling HPAI will depend on the poultry production system since the husbandry practices vary between production sectors.

2.2.5.1 Enhancing biosecurity measures

Biosecurity includes three major components: isolation, traffic control and sanitation (WHO 2006e). The use of biosecurity in poultry rearing according to FAO varies from high (closed, controlled heating and cooling system), to medium (open system, netting to prevent entrance of outside birds), to low (fences around poultry areas, poultry roam free in specified areas) to nonexistent (free ranging animals) (FAO 2006). In many resource-poor countries where the virus is or may be endemic, little or no biosecurity is employed in poultry farming (Desvaux et al. 2006; VSF 2004).

Improving biosecurity by restricting domestic and wild bird mixing, separating poultry areas from other domestic animal areas and separating poultry and human areas greatly reduces the likelihood of transmission between animals (FAO 2005; Kung et al. 2007). Also, practicing an “all in/all out” production system, which does not allow the introduction of new birds into the flock, and minimizing the number of people entering the farm or the amount of equipment shared between farms, reduces the potential of the virus from entering the farm either through infected animals or fomites (Meroz & Samberg 1995). Restricting poultry movement by fencing or caging animals is often difficult to implement in resource limited areas with free-ranging or organic farming because many farmers rely on animals to forage for their own food. It is also difficult to restrict poultry movement in international border areas because they are often open and uncontrolled.

2.2.5.2 Stamping out of infected and at-risk poultry

Mass culling has largely been successful at curbing transmission among commercial poultry in previous HPAI outbreaks in the Netherlands (H7N7) (Koopmans et al. 2004; Stegeman et al. 2004), Italy (H7N1) and Hong Kong (H5N1) (Capua & Marangon 2000; Capua et al. 2002; Ellis et al. 2004a; Mannelli et al. 2006). However, the economic costs associated with the loss of production and reduced livelihood are high (MOH 2007; Smith 2005).

Culling often focuses on the infected flock and uninfected, susceptible flocks (preemptive culling) located within a specific radius (usually between one or three km) around the infected farm (ring culling) or a larger area (e.g., all poultry in Hong Kong) (Rennie 2001). One modelling exercise suggested that immediate culling of infected flocks has greater efficacy in controlling viral spread than culling surrounding flocks (Le Menach et al. 2006). Another modelling exercise suggested that localized culling may have a limited impact on controlling viral spread and should include a larger radius around the infected premises (Truscott et al. 2007).

2.2.5.3 Controlling poultry movement

The economic forces driving the trade of animals and animal products have been shown to lead to wide spread and often uncontrolled/illegal movement of animals over large distances, particularly in regions of the world where movement is not regulated (Sims 2007). Non-regulated regions make movement exceedingly difficult to control. The movement of animals has played a key role in disease transmission for other animal disease outbreaks including the 2001 Foot and Mouth Disease (FMD) epidemics in the United Kingdom (Chis Ster & Ferguson 2007; Ferguson et al. 2001; Kao et al. 2006; Ortiz-Pelaez et al. 2006) and HPAI (H7N7) outbreaks in the Netherlands (Boender et al. 2007; Stegeman et al. 2004).

The connectedness of animal networks can lead to large and widespread epidemics of disease and an understanding of human and animal movement and their contact structures can be used to design more targeted surveillance activities and inform models of disease spread which could result in more cost-effective disease prevention and control (Colizza V et al. 2007; Dent et al. 2008; Green et al. 2008; Kiss et al. 2008; Truscott et al. 2007). For example, during the FMD outbreak in the UK, the movements of livestock facilitated the long-distance spread of the disease within a few months resulting in over 8.5 million animals slaughtered (Anderson 2002). The rapid analyses of livestock movements lead to the prompt implementation of control measures including restricting livestock movement that aided the control of the epidemic (Chis Ster & Ferguson 2007; Ferguson et al. 2001; Kao et al. 2006; Ortiz-Pelaez et al. 2006).

Poultry movement restrictions have been used within Europe, Asia and Africa following the detection of H5N1 (EU 2006; OIE 2006a; OIE 2009a). For example in England, following the detection of H5N1 in poultry in 2007, in addition to other outbreak control methods, a

three km “protection zone” was put in place around the location of the outbreak indicating that all poultry within the protection zone must be tested and kept indoors. In addition a ten km “surveillance zone” was put in place around the location of the outbreak prohibiting the movement of poultry to and from the area, restricted live bird markets in this zone, and placing footpath restrictions on free-range poultry farms within the zone (EU 2006). Upon detection of H5N1, other countries including Nigeria and Hong Kong restricted poultry movement outside their own country (OIE 2006a; OIE 2008c). Local movement restrictions are also common. For example, a temporary restriction of poultry trade outside the province was implemented following the most recent H5N1 outbreak in Cambodia (pers. comm. NaVRI).

Despite their likely role in the circulation and spread of HPAI in South East Asia, little is understood about the poultry market chains, legal or illegal trade of poultry or the types and frequencies of contact that exist between rural people raising poultry, local markets and large-national poultry markets in the major cities. Because trade of poultry may be responsible for some transmission of H5N1 within countries (Chen et al. 2005; Normile 2005a; WHO 2006-2009), controlling the movement of live poultry and poultry products could contain or reduce the spread of the virus. However, it is virtually impossible to restrict all illegal cross-border movement of poultry between countries with large and uncontrolled land borders (Sims 2007).

2.2.5.4 Live bird markets

Live bird markets (LBM) are common in Asian countries because of a cultural preference to consume freshly slaughtered meat (Webster 2004; Woo et al. 2006). The dense concentration of live birds and a high turn-over rate of birds (i.e., hosts) in these markets provide ample conditions for virus amplification (Webster 2004) and may be an important reservoir for HPAI or “hub” for circulation (Senne et al. 1992). Additionally, LBM are an ideal environment for transmission of avian influenza viruses from poultry-to-humans since they are frequented by large numbers of people (Woo et al. 2006).

It is unclear what role LBM has played in the circulation of HPAI/H5N1 in many Asian countries where LBM are prevalent. The close contact with live animals at such markets has been identified as a risk factor for SARS (Guan et al. 2003) and HPAI/H5N1 (Mounts et al. 1999). It has been demonstrated from investigations of past and current outbreaks and from HPAI surveillance programs in Vietnam, Thailand, Cambodia, China and Hong Kong, that

HPAI/H5N1 is circulating in the LBM (Amonsin et al. 2008; Guan et al. 2002; Kung et al. 2003a; Kung et al. 2007; MAFF Unpublished Data; Nguyen et al. 2005; Wang et al. 2006). It can also be assumed that HPAI/H5N1 may be circulating undetected in the markets of many other countries.

The movement of poultry through LBM has been shown to be an important factor in the circulation and spread of HPAI (Kung et al. 2007; Sims et al. 2003). In early 2002 in Hong Kong, an investigation into an outbreak first identified in LBM led to the discovery of the virus on rural farms that had sold chickens to the LBM (Sims et al. 2003). Further work determined that the contact between the retail market and chicken farms via humans was a significant risk factor for infection among chicken farms (Kung et al. 2007).

Control of avian influenza viruses within LBM focuses on implementing rest days, in which poultry stalls are emptied, cleaned and restocked. These efforts, which have been implemented in Hong Kong, have shown to reduce transmission of HPAI (H9N2) and other viruses among birds in LBM (Kung et al. 2003a).

2.2.5.5 Vaccination in poultry

There are several vaccines that have been used or are being developed to “prevent, manage or eradicate avian influenza.” The two most common vaccines used are oil-based emulsion inactivated whole low- or high-pathogenic virus vaccines and recombinant fowlpox virus-vector vaccines. Other vaccines include those based on reverse genetics or recombinant poultry vaccines made with HPAI and Newcastle diseases viruses (Swayne 2006; Swayne & Suarez 2007). When used correctly, vaccines have been shown to reduce infection rates, reduce morbidity and mortality rates, reduce viral shedding of the virus, and increase resistance to avian influenza virus infection by stimulating an immune response to the HA protein (Beato et al. 2007; Ellis et al. 2004b; Kim et al. 2008; Middleton et al. 2007; Peyre et al. 2009; Qiao et al.; Swayne 2006; Swayne et al. 2006; Swayne 2008; Tian et al. 2005; van der Goot et al. 2008; Webster et al. 2006b). Effective protection is conferred by the production of neutralizing antibodies of the NA protein and reached when clinical signs of disease are absent and mortality is prevented (Swayne 2008).

Vaccination can be implemented prophylactically, which could result in protective immunity in a susceptible population (usually applied in endemic situations), as a preventative measure for at-risk poultry, or as an emergency (reactive) measure during times of outbreaks or in

areas where widespread vaccination is impractical (Capua & Marangon 2006a; Capua & Marangon 2006b; EC 2006). Emergency vaccination, in which only high risk poultry are vaccinated, is used in combination with culling of infected and suspected poultry. The DIVA (Differentiating Infected from Vaccinated Animals) strategy uses sentinel birds (e.g., non-vaccinated) to monitor viral activity within the flock by placing unvaccinated sentinel birds on the same premises of vaccinated flocks to differentiate naturally infected from vaccinated birds (Capua & Marangon 2006a; Capua et al. 2003; EC 2006).

A number of experimental studies have been conducted to evaluate the effectiveness of HPAI vaccination in individual birds (Ellis et al. 2004b; Kim et al. 2008; Swayne et al. 2006; Tian et al. 2005; Webster et al. 2006b). Experimental studies have evaluated the protection provided by vaccines by challenging vaccinated birds with H5N1 and evaluating clinical signs, mortality, antibody response and viral shedding. These have demonstrated that clinical signs, mortality, and viral shedding are all reduced in vaccinated birds (Table 2-4 and Table 2-5) (Ellis et al. 2004b; Kim et al. 2008; Peyre et al. 2009; Swayne et al. 2006; Swayne 2008; Tian et al. 2005; Webster et al. 2006b).

Table 2-4 Summary of experimental studies of vaccine efficacy in chickens

Study	Vaccine type/ Challenge Strain	Study type (experimental or field)	Mortality	Morbidity	Viral Shedding	Antibody Response
Ellis et al 2004	Single dose killed inactivated H5N2 virus / H5N1 Hong Kong	Outbreak response in Hong Kong	Reduction at 9-18 dpv and mortality ceased at 18+ dpv	no evidence of clinical disease in vaccinated chickens	--	81.7% of chickens had H5 antibody titer \geq 16 by 22 dpv
Webster et al 2006	Single dose reverse genetics-derived vaccine containing HA-H5 and NA-H3 virus/ H5N1 Vietnam	Experimental	no mortality at 3, 5, 7, 10, 14 dpc	no clinical signs following vaccination	Tracheal shedding : 2/75 at 3 and 5 pdc; 0/75 at 10, 14 dpc Cloacal shedding (chickens): no shedding at 3,5,7,10, 14 dpc	100% had H5 antibody titer \geq 16 at 21 dpv
Tian et al 2005	H5N1 inactivated vaccine/ H5N1 China	Experimental	no mortality at 2,3, 43 dpv	no disease signs at 2,3,43 wpc	3/40 had low doses of oropharyngeal shedding 3 dpc no cloacal shedding at 3,5,7 dpc	--
Swayne et al 2006	H5N2 inactivated vaccine / H5N1 Indonesia	Experimental	90% protection from mortality	90-100% protection from clinical signs	Cloacal shedding present in 6/20 at 2 dpc oropharyngeal shedding in 11/20 at 2 dpc	100% had H5 antibody titer \geq 10 at 21 dpv
Qiao et al 2009	Recombinant fowlpox virus vector-based vaccine/ H5N1 China	Experimental	100% protection from mortality	--	no viral shedding in vaccinated birds at 4 dpc when challenged at 1, 2 and 40 wpv	Hi antibody titer >6.3 at 3 wpv

-- Not reported

dpv = days post vaccination; wpv = weeks post vaccination

dpc = days post challenge; wpc = weeks post challenge

Table 2-5 Summary of experimental studies of vaccine efficacy in ducks

Study	Vaccine type/ Challenge Strain	Study type (experimental or field)	Mortality	Morbidity	Viral Shedding	Antibody Response
Kim et al 2008	1 µg of HA protein in a single dose of inactivated oil emulsion whole-virus H5 influenza vaccines (3 different vaccines) / H5N1 Laos	Experimental	no mortality in vaccinated ducks; 90% of controls died	no disease signs in any of the vaccinated ducks	no viral shedding in vaccinated birds at 3, 5, 7 or 10 dpc	100% of ducks given 2 of the vaccines and 40% given the 3rd vaccine had H5 antibody titer ≥ 16 at 3 weeks p.v.
Beato et al., 2006	2 dose whole virus inactivated oil emulsion vaccine (H5N2 Potsdam)/ H5N1 Vietnam	Experimental	no mortality in vaccinated ducks	no disease signs in any of the vaccinated ducks	low levels of viral shedding at days 3, 5, 10 days post infection no viral shedding after 21 dpv	HI titers were significantly higher after second vaccination and detectable until day 135 of life
Middleton et al 2007	H5N3 reassortant virus vaccine and bivalent (H5N9 [Italy]+H7N1) vaccine/ H5N1 Vietnam	Experimental	no mortality at 6 wpv	no disease signs in any of the vaccinated ducks	no virus shedding at 5 and 7 dpc	no antibody response to challenge virus
Tian et al 2005	H5N1 inactivated vaccine/ H5N1 China	Experimental	no mortality at 3 wpv	no disease signs in any of the vaccinated ducks	no oropharyngeal or cloacal shedding at 3,5,7 dpc	--
Webster et al 2006	Single dose reverse genetics-derived vaccine containing HA-H5 and NA-H3 virus /H5N1 Vietnam	Experimental	no mortality at 3, 5, 7, 10, 14 dpc	no disease signs in any of the vaccinated ducks pc	no tracheal shedding at 3,5,10,17 dpc no cloacal shedding at 3,5,7,10, 14 dpc	100% of vaccinated chickens and ducks had H5 antibody titer ≥ 16 at 21 dpv; titers rise after re-vaccination
van der Goot et al., 2008	Single or double vaccination with Inactivated oil emulsion vaccine based on Mexico H5N2 / H5N1 China	Experimental	Mortality was reduced in vaccinated ducks	90% protection from clinical signs 7 dpv; 80% protection from clinical symptoms at 14 dpv	Tracheal shedding reduced after 7 dpv	HI titers with H5 antigen detectable in 2/20 ducks 7 dpc; in 3/20 ducks 14 dpc

-- Not reported

dpv = days post vaccination; wpv = weeks post vaccination

dpc = days post challenge; wpc = weeks post challenge

Vaccines against HPAI/H5N1 have been implemented in control programs in several countries including Hong Kong, Vietnam, Indonesia, China, India, Russia, Egypt, and Pakistan (Peyre et al. 2009; Swayne 2008). However, vaccine coverage and evaluation of vaccination effectiveness at the flock level is more difficult to conduct and there have been few studies published on the subject (Peyre et al. 2009). Vaccination is difficult to implement, especially in low-resource settings because currently available vaccines are administered by needle injection, often requiring multiple doses and commonly involving the direct handling of infected poultry. An ideal vaccine, which does not yet exist, would be a single dose vaccine given orally that is efficient for a number species and does not require refrigeration, especially relevant for developing countries with limited resources (Peyre et al. 2009).

In poultry, poorly implemented vaccination can mask the disease by reducing symptoms but not viral shedding, thus allowing the virus to circulate undetected and increasing the risk of antigenic shift (Tian et al. 2005). Vaccination coverage in Indonesia, for example, has not been universally applied throughout the country but implemented to strategically cover high risk regions of the country because of budgetary constraints and a limited vaccine supply. Although the vaccines available have been reported to be of “good quality,” they have not been “fully protective against some of the circulating strains in the country” (Siregar & Darminto 2008). Similarly, vaccine coverage in Vietnam has been incomplete and studies of vaccinated flocks suggest that protection is lower in southern areas of the country and that overall protection ranges from 28-55% (Nguyen 2008).

It is recommended that vaccination be used in conjunction with other HPAI control methods, including increased surveillance and detection (including both serologic and virologic testing), education for the public, culling of at risk poultry and enhanced biosecurity measures (Capua & Alexander 2006; Capua & Marangon 2006a; Capua & Marangon 2006b; EC 2006; Fasina et al. 2007; Guan et al. 2007; Peyre et al. 2009; Swayne 2006). Countries that have implemented a combination of control measures, including vaccination, restricting poultry movement, and culling of infected and susceptible poultry have been successful in controlling outbreaks of HPAI (Brown et al. 2007a; Ellis et al. 2004a; Henning et al. 2009; Tiensin et al. 2005). However, the results of these control measures are temporary as outbreaks continue to be reported (OIE 2009a). For example, in conjunction with other control measures, Vietnam introduced mass vaccination of over 100 million poultry in 2005 and experienced approximately one year without any outbreaks. However, in December 2006, several

outbreaks were reported and subsequently these have continued to be reported throughout 2007-2008, among unvaccinated flocks (ducks and geese) (Henning et al. 2009; Peyre et al. 2009).

As with any control strategy, there are advantages and disadvantages to vaccination. Table 2-6 outlines the major advantages and disadvantages for current control options for HPAI in poultry.

Table 2-6 Advantages and disadvantages of control options for HPAI in poultry

Control Methods	Advantages	Disadvantages
Increased biosecurity	Easy to implement in moderate, high income countries	<ul style="list-style-type: none"> • Difficult in settings with limited resources • Difficult in settings with free-ranging farms • Cost burden on farmer
Restricting movement	Effective in industrialised poultry settings	<ul style="list-style-type: none"> • Difficult to implement and monitor in limited resource settings
Culling	Fast, easy to implement	<ul style="list-style-type: none"> • Cost burden on farmer; compensation not always provided • Loss of potentially healthy flocks
Vaccination	Decreased mortality Decreased viral shedding	<ul style="list-style-type: none"> • Difficult to implement • Often requires multiple doses of vaccination • Time required for vaccine to become effective (e.g., 3 weeks) • May “mask” presence of disease in flocks; difficult to identify infection • Differences in immune response by species • Vaccine strain must be appropriate • Inappropriate/incomplete vaccination will not reduce viral shedding • Possible economic repercussions on farmers

2.3 Epidemiology and Transmission of HPAI/H5N1 in Humans

2.3.1 History of Influenza A Pandemics in Humans

New human pandemics arise when a novel NA subtype emerges amongst a susceptible human population with the ability for effective and sustained human-to-human transmission (Peiris et al. 2007). Table 2-7 below summarises the human pandemics of influenza A viruses over the last 150 years. The pandemic of 1918-1919 (H1N1) was particularly lethal in young, otherwise healthy adults, killing an estimated 40-50 million people worldwide (Horimoto & Kawaoka 2001; Hsieh et al. 2006; Kilbourne 2006; Webster et al. 1992). Genetic analyses of specimens collected from victims preserved in the arctic suggests that the strain was a novel

avian-like virus that adapted to humans (Taubenberger et al. 2005). The Asian Influenza Pandemic (H2N2) in 1957 and Hong Kong Influenza Pandemic (H3N2) in 1968 were less lethal and resulted from avian-human reassortment (Horimoto & Kawaoka 2001; Hsieh et al. 2006).

Table 2-7 Human pandemics and epidemics of the last 150 years

Year	Country of Origin	Serotype	Estimated Total Global Mortality (human)	Source/Emergence
1889	Europe? China?	H2N2	1-6 million	Unknown
1898	Europe	H3N2	500,000	Unknown
1917-1919	Europe? US?	H1N1	40 million	Novel avian-like virus
1957	Asia (China)	H2N2	6 million	Avian/Human reassortment
1968	Asia (China)	H3N2	2 million	Avian/Human reassortment

Source: (Gross 1996; Kilbourne 2006; Oxford 2000; WHO 2006-2009)

Since 1977 two subtypes (H1N1 and H3N2) have been circulating in humans worldwide. The isolation of H5N1 from a 3-year-old boy in Hong Kong in 1997 was the first occurrence of this novel strain in humans and signalled the emergence of a potentially new pandemic strain of avian influenza (Claas et al. 1998). H5N1 in Hong Kong in humans in 1997 did not emerge from reassortment; all of the genes found in this viral strain originated from an avian virus (Claas et al. 1998; Horimoto & Kawaoka 2001).

As of 30 December 2008, HPAI/H5N1 has infected 387 individuals in 15 countries (WHO 2006-2009). The number of cases is not evenly distributed throughout the world. By far, the largest number of human cases reported has been from Indonesia and Vietnam each having reported more than 100 cases (Table 2-8). No human cases have yet been reported in Western Europe or the Americas.

Table 2-8 reports the number of cases and fatalities in each country affected by H5N1 in humans, the clade or subclade that is circulating in the country and the median age and gender (% male) of the cases (WHO 2009b; Writing Committee of the Second World Health Organization Consultation on Clinical Aspects of Human Infection with Avian Influenza 2008). The overall case fatality rate (CFR) is 63.1% (median 62.5% IQR: 33.3-74.6) and varies by country (WHO 2009b).

Table 2-8 Case fatality rate of H5N1 in humans by country as of 30 December 2008

Country	Total		Case Fatality Rate (CFR) %	Clade or Subclade	Median age of cases (range)	% Male n/ total (%)
	Cases	Deaths				
Azerbaijan	8	5	62.5	2.2		
Turkey	12	4	33.3	2.2	16.5-10 (5-20) ^{‡‡}	9/16 (56) ^{‡‡}
Bangladesh	1	0	0	2.2	16 mo (--)	1/1 (100)
China	30	20	66.7	2.3	30 (12-41) [‡]	3/8 (38) [‡]
Djibouti	1	0	0	2.2	2 (--)	0/1 (0)
Egypt	50	22	44.0	2.2	12.5 (1-75) ^ª	12/38 (32) ^ª
Indonesia	137	112	81.8	2.1	18.5 (1.5-45) [‡]	33/54 (61) [‡]
Iraq	3	2	66.7	2.2	15 (3-39)	2/3 (66.7)
Lao People's Democratic Republic	2	2	100	2.3	28.5 (15-42)	0/2 (0)
Myanmar	1	0	0	NR	7 (--)	0/1 (0)
Nigeria	1	1	100	2.2	22 (--)	0/1 (0)
Pakistan	3	1	33.3	NR	25 (22-27)	3/3 (100)
Cambodia	7	7	100	1		
Thailand	25	17	68.0	1	14-22 (2-58) [†]	19/41 (46) [†]
Vietnam	106	52	49.1	1		
Cambodia ^{††}	8	7	85.7	1	16(3-28)	3/8 (37.5) ^{††}
Total	387	245	63.1	--	--	--

Adapted from (Biswas et al. 2008; WER 2008; WHO 2006-2009; Writing Committee of the Second World Health Organization Consultation on Clinical Aspects of Human Infection with Avian Influenza 2008)

†Data from 2004-2005 cases only

‡Data from 2005-2006 cases only

ªData from 2006-2007 cases only

‡‡Data from 2006 cases only

†† Data from all cases (n=8)

NR= Not released

2.3.2 Modes of Transmission of H5N1 in Human Populations

The investigations of human H5N1 outbreaks in the field—usually in rural locations of developing countries—are difficult to conduct and have often involved collection of only basic information about exposures. Thus data on exposure are typically limited to “recent contact with infected poultry” (WHO 2004) or the preparation of sick birds for consumption (WHO 2006d). The specific mode of transmission from exposure to infected poultry remains unknown and the lack of exposure information has limited our ability to evaluate risk factors for infection. In addition, the lack of large-scale seroprevalence studies in areas where H5N1 is recurrent has limited our understanding of the extent of infection in these countries.

Bird and Farrar have proposed a data collection form that details the minimum amount of information that should be collected from suspected human H5N1 cases, and which includes questions on direct and indirect contact with poultry and the timings of such contact (Bird & Farrar 2008). However, this questionnaire covers only general exposure information (e.g., handling sick or dead poultry, handling faeces or fertiliser from sick or dead poultry, slaughtering poultry) and does not include any potential transmission via the environment (e.g., contaminated water).

An illustration of known pathways of poultry-to-human infection of HPAI, particularly subtype H5N1, is shown in Figure 2-6. Transmission of H5N1 can occur via direct or indirect contact with an infected bird. Direct routes include contact with infected blood or bodily fluids via food preparation practices (Greiner et al. 2007) (e.g., slaughtering, boiling, defeathering, cutting meat, cleaning meat, removing and/or cleaning internal organs of poultry); ingesting undercooked or uncooked poultry products (e.g., eggs, meat, blood); through the care of poultry (either commercially or domestically); cleaning poultry cages or their designated areas; or using poultry faeces for fertilizer. Little is understood about H5N1 transmission via indirect routes, though recent studies have suggested an association between exposure to a contaminated environment (e.g., water) either through ingestion, conjunctival or intranasal inoculation of contaminated water (de Jong et al. 2005; Vong et al. 2008) or via fomites, such as shared equipment or vehicles transporting products between farms (FAO 2004a). Other pathways may exist but are currently unknown.

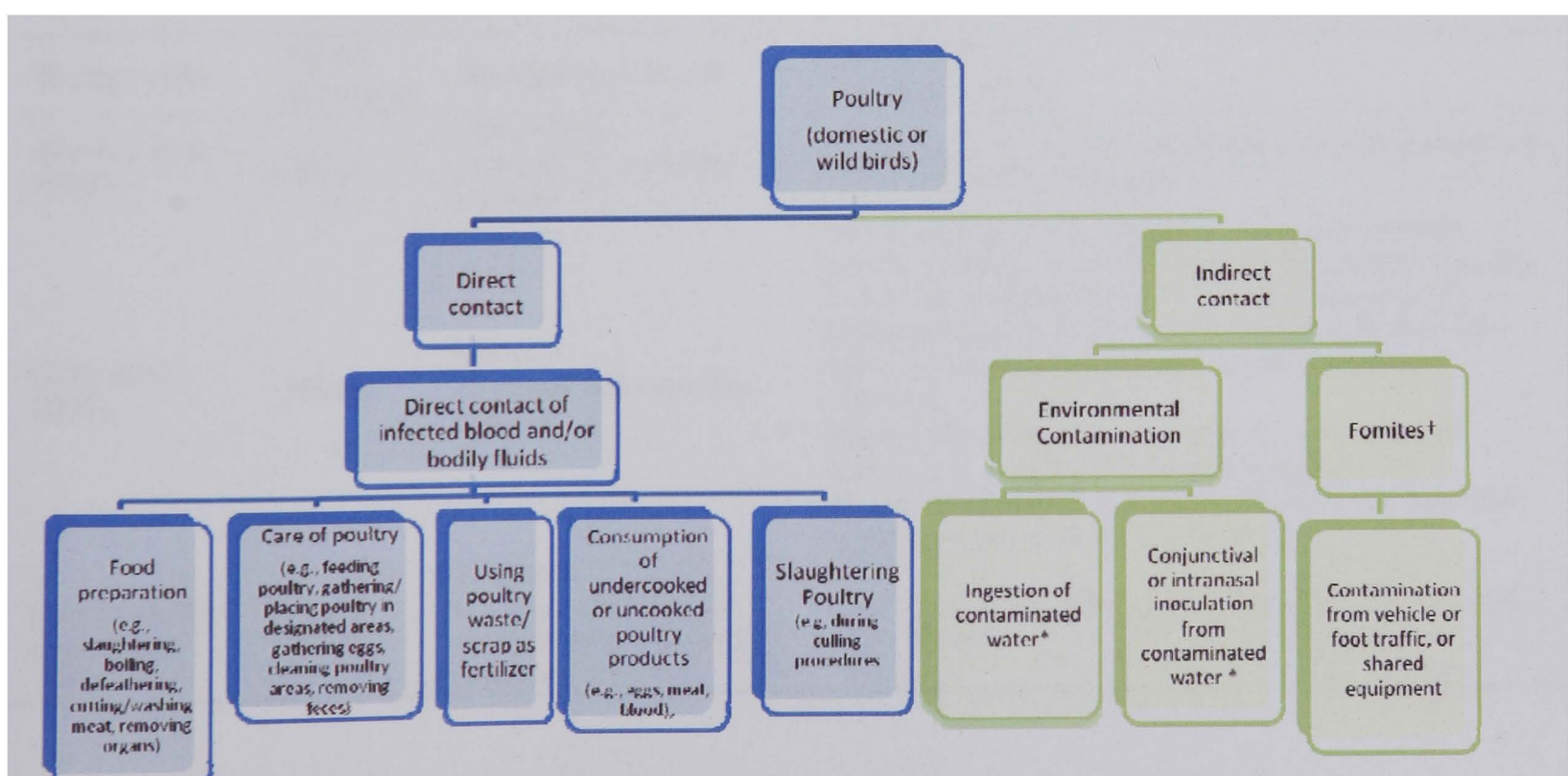


Figure 2-6 Known and suggested pathways to infection from poultry to humans

*via swimming/bathing in water frequently used by domestic and/or wild poultry

†Fomites are inanimate objects that can become contaminated and are capable of carrying infectious organisms

HPAI is transmissible from poultry-to-humans directly via contact with contaminated environments, through close contact with infected poultry or possibly through another animal species (e.g., pig, cat, dog, tiger) that serves as an intermediate host or mixing vessel (Alexander & Brown 2000; CDC 2005; Koopmans et al. 2004; Lipatov et al. 2008; Thanawongnuwech et al. 2005). Intimate contact with infected poultry (e.g., slaughtering, removing internal organs, licking wounds of bloody fighting cocks) is believed to be required for transmission of H5N1 from poultry to humans (Dinh et al. 2006; WHO 2006-2009). However, the extent of these behaviours is currently unknown and there is reluctance of individuals to disclose information on possible exposure from illegal activities. For example, an outbreak investigation in Azerbaijan in early 2006 found that the likely source of H5N1 in nine (eight confirmed, one probable) human cases was infected wild swans, with transmission probably occurring as a result of the illegal activity of de-feathering these birds (Gilsdorf et al. 2006).

A limited number of epidemiologic studies have been conducted throughout Asia and Africa to evaluate risk factors for human H5N1 infection. Most of these have been of a case-control design where they have evaluated exposure to poultry via visiting live poultry markets, through food preparation or caring or feeding poultry or contact with a confirmed human case. All of these studies, the results of which are summarized in Table 2-9, have included small numbers of subjects thus limiting the precision of their results.

Table 2-9 Risk factors for H5N1 infection: summary of published case-control studies

Study, year	HPAI Subtype	Study Population	Risk Factors RR, OR, 95%CI
(Mounts et al. 1999)	H5N1	Hong Kong 15 cases 41 matched controls	Exposure to poultry at live/wet markets was associated with a 4-fold increased risk Univariate Analysis: preparing/cooking unhealthy poultry (OR=31, 2.4-1150), having sick or dead poultry in the household (OR=7.41, 2.7-59), presence of sick/dead poultry in the neighborhood (OR=3.9, 1.0-55.7), no indoor water source in the household (OR=5.0, 1.3-77.0)
(Dinh et al. 2006)	H5N1	Vietnam 28 cases 106 matches controls	Multivariate Analysis: No water in the household (OR=6.5, 1.2-34.8), sick or dead poultry in the household (OR=4.9, 1.2-20.2), prepare and cook sick or dead poultry (OR=9.0, 0.98-82.0)
(Areechokchai et al. 2006)	H5N1	Thailand Matched case control study of 16 cases and 64 controls	Direct touching of unexpectedly dead poultry OR 29.0 (2.7--308.2)

2.3.2.1 Human Seroprevalence Studies

To date, a few small-scale human seroprevalence studies have been conducted in Hong Kong, China, Thailand, Nigeria, Cambodia, and Vietnam to determine the frequency of asymptomatic or subclinical infection and evaluate risk factors for HPAI/H5N1 virus infection (Apisarnthanarak et al. 2005; Bridges et al. 2000; Bridges et al. 2002; Katz et al. 1999; Ortiz et al. 2007; Schultsz et al. 2005; Thanh Liem et al. 2005; Vong et al. 2006; Vong et al. 2009). These studies are summarized in Table 2-10 and can be categorized by the study populations evaluated in each study: occupationally exposed individuals (health care workers or poultry workers) or non-poultry related occupational settings (subjects living or working in close proximity to confirmed H5N1 case):

Occupationally exposed persons: poultry workers

The following three studies evaluated the frequency of asymptomatic or subclinical infection and evaluate poultry-to-human risk factors for H5N1 and H7N1 virus infection among poultry workers:

- *Bridges et al 2002*: The risk of H5N1 was evaluated among poultry workers involved in the culling of all poultry in Hong Kong following the first reported human H5N1 case in a child in Hong Kong in 1997. Among the 1525 poultry workers and 293 government workers enrolled, 83 (5.3%) poultry workers and nine (3.1%) government workers tested positive for H5N1 antibodies by both microneutralisation and Western Blot.

A nested case-control study evaluated the risk factors for infection among the poultry workers (n=81) when compared to unmatched controls. Risk factors associated with infection included work in retail vs. wholesale/hatchery/farm/other poultry industry OR=2.7 (95% CI 1.5-4.9); >10% mortality among poultry with which they had worked in the previous two months OR=2.2 (95% CI 1.3-3.7); butchering poultry OR=3.1 (95% CI 1.6-5.9); feeding poultry OR=2.4 (95% CI 1.4-4.1); and preparing poultry for restaurants OR=1.7 (95% CI 1.1-2.7). The study found that subjects exposed to intense contact with poultry during the culling processes were at an increased risk for infection with H5N1. It also found that exposure through trading poultry at retail markets was associated with increased risk of H5N1 infection.

- *Ortiz et al 2007*: Upon confirmation of a H5N1 outbreak in poultry in Nigeria in 2006, the risk of H5N1 infection among poultry workers and laboratory workers in contact with H5N1 was evaluated. Two-hundred and ninety-five poultry workers who had been exposed to infected poultry occupationally and domestically participated in the study. Home exposure to poultry included owning any (54%) or sick poultry (42%) or touching live or dead poultry (81%). None of the 295 poultry workers or 25 laboratory workers tested positive for H5N1 antibodies by microneutralization and HI assay using horse red blood cells. This study found no evidence of poultry-to-human transmission among poultry and laboratory workers in contact with infected poultry.
- *Puzelli et al 2005*: The risk of HPAI/H7N1 and LPAI/H7N3 was evaluated among Italian poultry workers of farms affected by an outbreak of HPAI/H7N1 between 1999 and 2003. No serum samples tested positive for HPAI/H7N1 (0/672).

Occupationally exposed persons: health care workers

The following four studies evaluated the frequency of asymptomatic or subclinical infection and evaluated human-to-human transmission risk factors for H5N1 virus infection among health care workers:

- *Bridges et al 2000*: The risk of H5N1 among health care workers involved in the care of confirmed H5N1 patients in Hong Kong in 1997 was compared to health care workers without known exposure to confirmed cases but with similar patient responsibilities. Because diagnosis was delayed, infection control procedures were not immediately initiated. Risk factor data were collected on exposure to the case patient (provided direct care to case, physical contact, face-to-face talking, worked within two metres of patients, recalled patient coughing/sneezing, suctioned respiratory secretions from or administered breathing treatments to patients, changed bed linens or bathed patient), age, sex, occupation and exposure to poultry (shopped at live poultry market, had live or freshly cut poultry in their home in the weeks before interview).

Among the exposed and unexposed health care workers enrolled, 4% (8/217) and 0.7% (2/309) tested positive for H5N1 antibodies using microneutralisation and Western Blot techniques, respectively. Risk factors for infection included changing bed linens (no OR provided) and did not include exposure to poultry (no results

provided). It was suggested by the authors that the severity of illness in the H5N1 patients may have been more likely to result in transmission from humans-to-humans because of higher viral concentrations found in respiratory specimens.

- *Apisarnthanarak et al 2005*: Occupational exposure to H5N1 of 49 health care workers exposed to a confirmed H5N1 patient in a university hospital setting in Thailand was evaluated in a seroprevalence study. Health care workers were classified as exposed (n=25) and non-exposed (n=24) to the patient and did not differ by demographic characteristics or exposure to poultry (contact with ill poultry, shopping at live poultry market, had live or freshly cut poultry in their home in the two weeks before interview or history of living on a poultry farm). The use of personal protective equipment (PPE, surgical mask, gown and gloves) was not initiated until 48 hours after the case was admitted to the hospital. No health care workers tested positive for H5N1 antibodies using microneutralisation and Western Blot techniques and thus there was no evidence of person-to-person transmission of H5N1 in this study.
- *Schultsz et al 2005*: Occupational exposure to H5N1 was evaluated among health care workers exposed to confirmed H5N1 patients in a Ho Chi Minh City hospital, Vietnam. None of the 60 health care workers involved in the care of H5N1 patients tested positive for H5N1 antibodies using ELISA or microneutralisation and Western Blot techniques despite 25.4% having reported contact with the patients secretions, approximately half (29/59) reporting to have spent >12 hours with the patient and limited use of control measures or personal protective equipment (e.g., gloves). No evidence of human-to-human or poultry-to-human transmission of H5N1 occurred among health care workers.
- *Thanh Lim et al 2005*: Occupational exposure to H5N1 of health care workers exposed to four confirmed and one probable H5N1 patients in a Hanoi hospital was evaluated in a seroprevalence study. None of the 83 health care workers who provided a single blood sample and completed a questionnaire to obtain information on demographic characteristics, medical history, use of protective equipment while in contact with the case, exposure to the cases, or exposure to poultry tested positive for H5N1 antibodies using microneutralisation and Western Blot techniques.

The use of PPE was high among subjects with 94.8% reporting that they always wore a mask while examining or caring for H5N1 patients, while 31.6% reported that they always wore eye protection, 61.5% reported that they always wore gloves while in contact with H5N1 patients.

Non-occupational exposure: household and social contacts

The following four studies evaluated the frequency of asymptomatic or subclinical infection and evaluate poultry-to-human risk factors for HPAI/H5N1 virus infection among subjects living or working in close proximity to confirmed H5N1 outbreaks in human and domestic poultry populations:

- *Katz et al 1999*: The frequency of asymptomatic or sub-clinical H5N1 infection was evaluated among household or social contacts of 17 confirmed human H5N1 cases in Hong Kong. Six of the 51 household contacts and none of the 26 social contacts (26 social contacts who participated in a 4 day tour with one case plus 23 co-workers) tested positive for H5N1 antibodies using microneutralisation and Western Blot techniques. Although not statistically significant, the authors suggest that exposure to poultry in their homes was a likely risk factor for infection (21% of seropositive subjects had contact with poultry versus 5% of seropositive subjects with no poultry contact, $p=0.13$).
- *Vong et al 2006*: The frequency of asymptomatic or sub-clinical H5N1 infection was evaluated among residents living within a 1km radius where a man was confirmed with H5N1 infection in Cambodia. Three-hundred and fifty one subjects were recruited in the study; however none tested positive for H5N1 antibodies using microneutralisation and Western Blot techniques despite frequent contact with poultry and 96 of 262 (36.6%) households with probable H5N1 infection in chickens.
- *Vong et al 2009*: The frequency of asymptomatic or sub-clinical H5N1 infection was evaluated among residents living within a 1km radius of two human H5N1 cases in two rural villages in Cambodia. Among the 674 subjects recruited, seven (1.0%) tested positive for H5N1 antibodies by microneutralisation and Western Blot. All seven cases were ≤ 18 years old and six of the seven were male (85.7%). Risk factors for infection—including handling poultry, practices involved in the preparation of food, contact with confirmed cases, hand hygiene after contact with poultry and

general health—were evaluated in a retrospective matched case-control study of the seven subjects and 24 matched controls (for sex, age [± 3 yrs], village of residence and households with H5N1).

Risk factors associated with testing positive for H5N1 antibodies included swimming or bathing in ponds OR=11.3 (95% CI 1.25-102.18) and gathering poultry and placing them in cages or designated areas OR=5.8 (95% CI 0.98-34.12). These results taken in conjunction with recent evidence of H5N1 virus in the surrounding areas where poultry died from H5N1 infection (Vong et al. 2008) indicate that swimming or bathing in ponds located around the household where poultry typically have access may be a risk factor for infection. It is worth noting that one case had only spent five days in the village during the study period (approximately three months) and had reported preparing poultry for consumption and cleaning poultry faeces in his house yard during that 5-day period.

- *Hinjoy et al, 2008*: A seroprevalence studies in rural Thailand (Hinjoy et al. 2008) was conducted to evaluate asymptomatic infection among poultry farmers in rural areas where H5N1 outbreaks had been confirmed. No farmers in rural Thailand (n=322) farmers tested positive for anti-H5 antibodies by microneutralisation and Western Blot techniques.

Table 2-10 Results of seroprevalence studies to determine the frequency of asymptomatic or subclinical infection of H5N1 virus

Reference	Study Population & Year of Outbreak	Transmission Evaluated	Seroprevalence Results (% seropositive)	Risk Factors RR, OR, 95%CI	Comments
Occupationally Exposed Persons: Poultry Workers					
(Bridges et al. 2002)	Poultry workers, Hong Kong 1997	Poultry-to-humans	9/293 (3%) government workers were seropositive 81/1525 (5.3%) poultry workers tested positive for H5N1 antibodies Nested case-control study conducted among 81 cases and 1231 controls	Work in retail vs. wholesale/hatchery/farm/other poultry industry 2.7 (1.5-4.9) >10% mortality among poultry 2.2 (1.3-3.7) Jobs: · Butchering poultry 3.1 (1.6-5.9) · Feeding poultry 2.4 (1.4-4.1) · Handling money 1.6 (1.0-2.5) · Preparing poultry for restaurants 1.7 (1.1-2.7) Controlled for age-group	Limited poultry-to-human transmission among poultry and government workers involved in poultry culling operations
(Ortiz et al. 2007)	Poultry workers, Kano Nigeria 2006	Poultry-to-humans	0/295 poultry workers with median 14 days exposure to H5N1 0/25 laboratory workers with exposure to H5N1	None	No evidence of H5N1 infection with subjects with repeated exposure to infected poultry
Occupationally Exposed Persons: Health Care Workers					
(Bridges et al. 2000)	Health care workers, Hong Kong 1997	Human-to-human; poultry-to-human	10/526 (8/21 exposed; 2/309 non exposed HCW)	Changing the bed linen of cases (no OR provided); controlled for poultry exposure	Limited human-to-human transmission
(Apisarnthanarak et al. 2005)	Health care workers, Thailand 2004	Human-to-human; poultry-to-human	0/25 among health care workers in direct contact with H5N1 patient	None	No serologic evidence of H5N1 among health care workers with direct contact with human H5N1 patient
(Thanh Liem et al. 2005)	Health care workers, Vietnam 2004	Human-to-human; poultry-to-human	0/83 among health care workers, 95% of which had direct contact with confirmed H5N1 patients	None	No serologic evidence of H5N1 among health care workers with direct contact with human H5N1 patient
(Schultsz et al. 2005)	Health care workers, Vietnam 2004	Human-to-human; poultry-to-human	0/60 healthcare workers in contact with confirmed H5N1 patients	None	No serologic evidence of H5N1 among health care workers with direct contact with human H5N1 patient

Reference	Study Population & Year of Outbreak	Transmission Evaluated	Seroprevalence Results (% seropositive)	Risk Factors RR, OR, 95%CI	Comments
Non-Occupational Exposure: Household and Social Contacts					
(Katz et al. 1999)	Household and Social contacts of H5N1 patients, Hong Kong 1997	Human-to-human; poultry-to-human	6/51 (12%) household contacts 0/47 co-workers tested positive for H5 antibodies	None significant; however 21% of seropositive had contact to poultry vs. 5% of seropositive with no poultry contact, p=0.13	Human-to-human transmission was limited
(Vong et al. 2006)	Rural villagers living in the same villages as two confirmed H5N1 human cases 2005	Poultry-to-human	0/351 villagers tested positive for H5N1 antibodies	None	No evidence of H5N1 infection among subjects living in villages with conformed H5N1 in domestic poultry flocks; poultry-to-human transmission was low in this setting
(Vong et al. 2009)	Rural villagers living in the same villages as confirmed H5N1 human case 2006	Poultry-to-human	7/674 (1%) seropositive for H5N1 antibodies ≥1:80 85.7% (6/7) male All ≤18 years old Matched case-control study conducted with 7 cases and 24 controls	Swim/bathe in ponds OR 11.3 (1.25-102.2) Water source 6.8 (0.68-66.4) Gathered poultry and placed in cages or designated areas 5.8 (0.98-34.1) Removed/cleaned faeces from cages or poultry areas 5.0 (0.69-36.3)	Poultry-to-human transmission was low; possible transmission from the environment to humans via contaminated water
(Hinjoy et al. 2008)	Rural poultry farmers in Thailand, 2004	Poultry-to-human	0/322 farmers tested positive for H5N1 antibodies	None	No evidence of H5N1 infection among subjects living in villages with conformed H5N1 in domestic poultry flocks

PPE = personal protective equipment including masks, gloves, eye protection

2.3.2.2 Clusters of H5N1 in Humans

Suspected clusters of epidemiologic linked H5N1 cases have occurred among blood relatives in several countries, including Indonesia, China, Turkey, Azerbaijan, Vietnam and Thailand, suggesting that human-to-human transmission between family members may have occurred (Gilsdorf et al. 2006; Kandun et al. 2008; Kandun et al. 2006; Olsen et al. 2005; Oner et al. 2006; Ungchusak et al. 2005; Wang et al. 2008). An early investigation in Vietnam, suggested that between January 2004 and July 2005, 15 suspected family clusters occurred among the first 109 cases, of which nine clusters had at least two laboratory confirmed H5N1 cases. These results suggested that in three of the laboratory confirmed clusters exposure to a common source was unlikely as the timing of infections exceeded one week of each other (Olsen et al. 2005).

A family cluster in mainland China occurred in a father and son, the former likely infected through close, unprotected contact via care at a hospital of his son during his illness (Wang et al. 2008). Similarly in Thailand, a mother and aunt of an infected patient likely became infected through unprotected hospital care of their daughter/niece (Ungchusak et al. 2005). In Turkey, several members of the same family became infected with H5N1, however transmission likely occurred from poultry-to-human rather than human-to-human since they all shared the same living space with poultry (Oner et al. 2006).

In Indonesia, there have been 11 clusters of H5N1 among blood relatives with each cluster involving 2-7 blood relatives (Kandun et al. 2008; Kandun et al. 2006). Among the first three clusters, which occurred in 2005, limited human-to-human transmission may have occurred in two of the three clusters. Exposure to the virus via a contaminated environment, through contact with contaminated manure or with infected poultry could not be ruled out (Kandun et al. 2006). In a detailed analysis of all human H5N1 cases in Indonesia, the authors examined direct and indirect exposure to poultry and could not rule out a common source of infection in the clusters since families may have similar opportunities for exposure to the virus. While there may have been limited human-to-human transmission in some clusters, the authors suggest that genetic variation in families could result in the occurrence of clusters because of a predisposition to infection (Kandun et al. 2008). Cluster investigations have suggested that

some individuals may be more genetically susceptible to infection, Interpretations of the family clusters are often difficult because all of the suspected patients may not have been tested for H5N1.

2.3.3 Indirect-Transmission of H5N1 to Humans

It is possible for HPAI/H5N1 to be transmitted to humans indirectly via contact with fomites or through the environment (de Jong et al. 2005; FAO 2004a; Vong et al. 2008; Vong et al. 2009; WHO 2006g). Since birds are known to shed high concentrations of virus into water sources, transmission from poultry-to-humans through contaminated water is possible (WHO 2006g). The epidemiologic investigation of two H5N1 related cases in Vietnam suggested that exposure to possibly contaminated canal water via swimming or washing may have played a role in the acquisition of infection. However, the role of water in transmission could not be confirmed nor extrapolated since no further follow-up studies were conducted (de Jong et al. 2005). More recently, results from environmental sampling within a village with confirmed H5N1 in domestic poultry flocks and one human case as well as results from a human seroprevalence study from the same villages in Cambodia identified contaminated water as a potential risk factor for H5N1 infection (Vong et al. 2008; Vong et al. 2009).

2.3.4 Summary of Epidemiology Studies

Epidemiologic studies of H5N1 in humans have identified several risk factors for infection including close contact with poultry and transmission via the environment. However, despite frequent and widespread contact with poultry, transmission from poultry to humans is rare. A small number of cases resulting from human-to-human transmission are likely to have occurred among blood relatives. However poultry exposure could not completely be ruled out in some cases. It is likely that there are genetic and/or immunological factors that render some more susceptible to infection than others.

Several important data gaps currently limit our understanding of the epidemiology of H5N1 in humans. There remains considerable scope for underreporting of human cases. We currently lack sufficient exposure data from the confirmed H5N1 cases around the world to fully evaluate other potential risk factors (e.g., the environment) for infection. The seroprevalence

studies that have evaluated the frequency of asymptomatic or subclinical infection and risk factors for H5N1 infection have identified few asymptomatic individuals with anti-H5N1 antibodies, indicating previous infection with H5N1. However, it is not possible to determine whether this is a true reflection of HPAI/H5N1 infection given the limited geographical scope of such studies to date. There may be other factors that limit transmission to humans including differences by age in intrinsic susceptibility to infection, pre-existing cross immunity arising from previous exposure to other human influenza A virus and/or clinical presentation of disease.

2.4 Review of HPAI/H5N1 in Cambodia Prior to 2006

The remainder of this chapter describes HPAI/H5N1 research carried out in Cambodia prior to the start of my field work in April 2006. The first few months of my field work in Cambodia were spent identifying, obtaining and reviewing all available research conducted on H5N1 in the country in order to evaluate the research carried out to date and to develop research questions that would address data gaps.

Cambodia is located in South East Asia, sharing borders with Thailand, Laos and Vietnam, and is administratively split into 24 provinces. Two major rivers, the Mekong and Tonle Sap, bisect the country. The population is approximately 14.2 million, 80% of which live in rural areas of the country (Figure 2-7). Agriculture (mainly rice farming), fisheries and forestry are the dominant industries. Temperature varies very little during the dry (December to April) and rainy (May to November) seasons.



Figure 2-7 Population density in Cambodia

Map generated using data from a Columbia University population study done originally in 2005 and updated in 2006, and is a 2010 projection (pers. Comm.)

2.4.1 Data on HPAI/H5N1 in Cambodia

2.4.1.1 HPAI/H5N1 in poultry in Cambodia

Data on H5N1 in poultry is available in the form of outbreak investigations, HPAI surveillance activities, and knowledge attitudes and practice (KAP) surveys (Table 2-11). The Food and Agriculture Organization (FAO), along with National Veterinary Research Institute (NaVRI), have funded enhanced passive surveillance systems of domestic poultry in several provinces using village animal health workers (VAHW) who are trained to identify and report acute high mortality in chicken (>60% mortality) and duck flocks (>30%).

Table 2-11 Summary of existing data on H5N1 in Cambodia in 2006

Type of Study	Data Source	Description of data	Location	Date
Outbreak investigations:				
Backyard poultry (ducks/chickens)	IPC	Multiple outbreaks (~20 confirmed H5N1) Questionnaire/ Serology	PV, KgC, Kampot, Kg Speu	Multiple dates
Ducks/chickens/ fighting cocks	IPC	KAP/ Serology, Ta Sen village, So Tip commune, Choeung Prey district	KgC	Feb 06
HPAI surveillance programme in Cambodia (Desvaux et al. 2006)	MAFF	Poultry density by province	All	2002
Poultry ownership and handling behaviour(Ly et al. 2007)	IPC	KAP Survey in 25 villages	PV, KgC	Jan '06
Sentinel duck surveillance survey	IPC	250 of 333 (76%) households surveyed, 16% of households raised ducks; 80% duck flocks free ranging	PV	Oct 5-8, '05
Village animal health Worker training	MAFF	Active surveillance system of poultry mortality using trained paraprofessionals	KgC, PV, Kg Speu*	Ongoing
Impact on HPAI on poultry producers	VSF	Survey of poultry producers production	All	2004
Environmental sampling	IPC	Approx. 30 water, soil samples per outbreak area	Kg Cham, PV	2006 & ongoing

*each month, new villages are included in training of VAHW

PV= Prey Veng, KgC = Kampong Cham; Kg Speu = Kampong Speu; All = entire country

2.4.1.2 Confirmed H5N1 infection in poultry

Outbreaks of H5N1 in poultry were first reported in poultry in early 2004 (Desvaux et al. 2006; OIE 2009a) and since then, at least 20-25 outbreaks have been confirmed in Cambodia (Figure 2-8) (MAFF Unpublished Data; OIE 2009a). HPAI/H5N1 mortality has been high in infected chicken flocks, often exceeding 90%, whereas HPAI/H5N1 mortality within duck flocks has ranged from relatively low mortality (<30%) to as high as 80-90%.

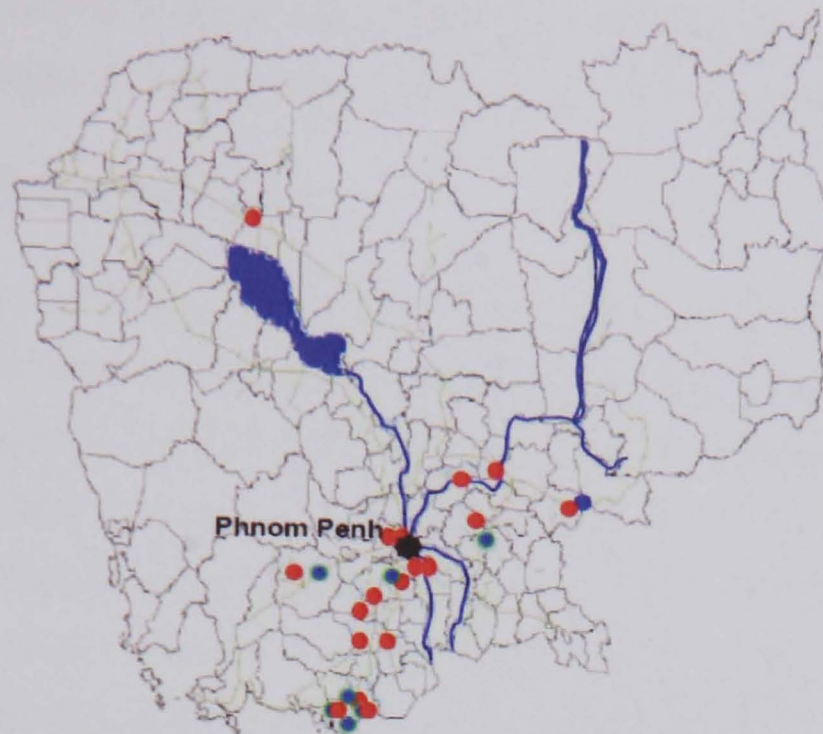


Figure 2-8 Locations of confirmed H5N1 outbreaks in poultry and humans

Locations of poultry outbreaks identified by blue dots; human outbreaks by red dots

2.4.1.3 Sentinel duck surveillance

In October 2005, a sentinel duck surveillance program was started in three villages near Kdey Boun Lake in Kampong Cham Province. In each village, 10 ducks from three flocks (300 ducks in total) were tagged and sampled (blood; cloacal and tracheal swabs) bimonthly. One month into the program (i.e., the second sampling), however H5N1 was isolated from one flock and as a result all duck flocks in the villages were subsequently culled and the program was stopped. Any mortality occurred in 54% of flocks and occurred in young populations (median age at death 3 months; range 1-24 months).

2.4.1.4 Environmental sampling

IPC has been actively involved in collecting environmental samples from villages with confirmed poultry H5N1 infection and has recently published their findings (Vong et al. 2008). In a survey conducted in a village where the 5th, 6th and 7th human H5N1 cases occurred, a total of 167 environmental samples (including soil, mud, faeces, soil swab specimens, swabs of feathers from poultry that recently died, and water plants around the household) were collected from 43 households. Twenty seven of 77 samples tested positive for H5N1 by RT-PCR and

viral RNA was found in 50% of poultry faeces samples, 50% of soil swab specimens samples, 50% of water plants samples, 50% of swabs from feathers and 29% of mud samples.

2.4.2 Research in Human Populations

Since 2005 there have been eight human cases (87.5% fatality) of H5N1 in Cambodia. However it is likely that some human cases have gone undetected (Normile 2005b; WHO 2006a). All cases occurred in individuals that lived in southern Cambodia (4 had occurred prior to the initiation of my PhD; Figure 2-8) and all had possible exposure to sick/dead poultry (WHO 2006b; WHO 2006c). The majority of cases were female (37.5% male) and the median age of the cases is 16 years old (range 3-28). Prior to the initiation of my PhD work, research on H5N1 in human populations had been conducted in three ways: KAP surveys, outbreak investigations and seroprevalence studies. Table 2-12 summarises studies undertaken since January 2004.

Table 2-12 Summary of existing data on H5N1 in human populations in Cambodia in 2006

Type of data	Data Source	Description of data	Location(s)	Date	Serology (Y/N)
Outbreak investigations	MOH, IPC, WHO	Serology of cases and close contacts; questionnaire of exposure to poultry, occurrence of poultry mortality	Kampot, PV, Kg Speu	Within 1 day of PCR confirmation from IPC	Y
Influenza-like Illness (ILI) Surveillance System	WHO, MOH, IPC,	Purpose is to understand epidemiology of influenza in Cambodia, help the MoH target control measures and detect outbreaks early	Takeo, Battambang, Siem Reap, Kg Cham, Phnom Penh	Start Aug '06	Y
Seroprevalence surveys (Vong et al. 2006; Vong et al. 2009)	IPC	Three cross-sectional antibody seroprevalence survey (n=1,025)	Kampot, PV, Kg Speu	May '06	Y
KAP Surveys(Ly et al. 2007)	IPC, WHO, NAPHC	Poultry ownership and handling behaviour in backyard poultry owners	PV, Kg Speu, Kg Cham, Kampot	Jan '06	N

PV= Prey Veng, Kg Cham= Kampong Cham; Kg Speu= Kampong Speu

2.4.2.1 Transmission from poultry to humans

Seroprevalence studies have been conducted since 2005 in villages where human cases occurred (Kampot 2005, Prey Veng and Kampong Speu 2006). These have been described in the previous section (Vong et al. 2006; Vong et al. 2009).

2.4.2.2 KAP survey of rural Cambodians January 2006

A knowledge-attitudes-practice (KAP) study was conducted in Kampong Cham and Prey Veng provinces to determine the extent of exposure to backyard animals and to obtain an in-depth understanding of the behaviour of people domestically exposed to poultry (Ly et al. 2007). The results confirmed previous data showing that poultry ownership is high (97% and 39% owned chickens and ducks, respectively), but flock size is small (median flock size ten and six for chickens and ducks, respectively). Species mixing was common and almost all flocks were free ranging (100% and 96% for chickens and ducks, respectively). In the six months prior to sampling, 60% of households (n=269) experienced any poultry mortality, however only 7% reported this to authorities. The sampling population used in this survey only included adult farmers (n= 460), thus limiting the comparisons that could be made across age groups and occupations.

2.4.3 Control and Containment of HPAI in Cambodia

Control of HPAI/H5N1 in poultry in Cambodia is managed by national and provincial veterinarians from the National Veterinary Research Institute (NaVRI), which is part of the Ministry of Agriculture, Fisheries and Forestry (MAFF) and Department of Animal Health and Production (DAHP). A national campaign to educate rural farmers in avian influenza symptoms in poultry and ways to protect against infection began in 2006 and continues into 2009. This program trains village animal health workers (VAHW), who are lay persons living in rural villages, to recognize and report high mortality in poultry. The training teaches VAHW to recognize high mortality in chicken (>60% over 1-2 days) and/or duck (>30%) flocks and inform the Provincial level health department, who in turn informs NaVRI of the potentially infected flock. Duck and chicken samples (blood, cloacal, tracheal) are taken from the infected flock and tested using egg inoculation (for the presence of influenza A, subtype H5 virus) at NaVRI in Phnom Penh. Positive samples are confirmed by RT-PCR at IPC, Cambodia's National Influenza Centre.

Upon confirmation, NaVRI conducts a formal outbreak investigation (retrospective poultry mortality survey) within a one km radius of the infected flock. A questionnaire assesses poultry ownership and mortality and blood, cloacal and tracheal samples are collected from a convenient selection of ten ducks and any noticeably sick chickens from each flock within the 1km radius. Culling of all poultry within a one km radius will commence only after confirmation of H5N1 by RT-PCR, typically 2-3 days after infection in the flock/village is detected (Figure 2-9). Vaccination has not been introduced in Cambodia and the government does not offer compensation for culled flocks.

In December 2008, H5N1 was reported in Cambodia's 8th human case. Follow-up investigations by NaVRI found H5N1 in poultry in the village in Kandal where this case lived and subsequently culled 300+ chickens and ducks. Control procedures also included movement restrictions on poultry from Kandal Province for three months.



Figure 2-9 Culling operations in Cambodia following detection of H5N1 in poultry

These photographs were taken during the culling operations following confirmation of H5N1 in poultry in Prey Veng Province in August 2006. The photograph on the bottom right illustrates an example of the environmental sampling that takes place during outbreak investigations.

2.5 Review of Research Questions

As stated in Chapter 1, the overall aim of this thesis is to evaluate poultry movement and the extent of interaction between humans and poultry in Cambodia to better define the risks of sustained transmission of H5N1 in poultry and onward potential transmission to humans. Two research questions (below) were designed to address gaps in knowledge of HPAI/H5N1:

Research Question 1: What is the frequency and extent of exposure to poultry in the general as well as occupationally exposed populations in Cambodia?

Research Question 2: How do current movements of poultry influence the potential spread of HPAI at local, regional and national levels? What are the implications of these movements for control and containment of H5N1 in poultry and/or human populations?

Chapter 3 Cross-Sectional Survey of Rural Cambodians: Study Methods and Subject Characteristics

3.1 Introduction

As described in Chapter 2, Cambodia is located in SE Asia sharing borders with Thailand, Laos and Vietnam. The capital of Cambodia is Phnom Penh and the Mekong and Tonle Sap rivers bisect the country intersecting in the capital. Temperature varies little during the dry (December to April) and rainy (May to November) seasons. The population is approximately 14.2 million and approximately 84.3% of the population lives in rural areas (NIS 2002). Life expectancy in Cambodia is approximately 60 years for men and 64 years for women (CIA 2008).

Cambodia is administratively split into 24 provinces (20 provinces and 4 municipalities, including Phnom Penh), 183 districts, 1,609 communes and 13,406 villages. The country has a democratic government under a constitutional monarchy and is composed of three sides of government: the Judiciary; the National Assembly, which holds legislative powers; and the Royal Government, which is composed of a Council of Ministers (25 Ministries) and is headed by Prime Minister.

The National Veterinary Research Institut (NaVRI) in the Department of Animal Health and Production (DAHP), which is a department within the Ministry of Agriculture, Fisheries and Forestry (MAFF), is responsible for all policies and planning regarding the health of animals in Cambodia. The DAHP is composed mainly of veterinarians that are stationed in Phnom Penh, and have offices in all province and some district centres. DAHP staff are responsible for monitoring the health of animals within their province and/or district geographic boundaries. This research was conducted in collaboration with the DAHP.

At the time this PhD proposal was designed, only a limited number of seroprevalence studies had been conducted and although it was hypothesized that recent contact with infected poultry was the main transmission route from poultry to humans, the specific mode of transmission

from animal to human was unknown (Bridges et al. 2002; Katz et al. 1999; Mounts et al. 1999; Vong et al. 2006). Previous surveys in Cambodia determined that domestic poultry ownership is high in rural areas of Cambodia (Ly et al. 2007). However none evaluated the current extent of exposure to poultry among the population. Therefore a large-scale cross-sectional survey of adults and children living in rural areas throughout Cambodia was designed to evaluate poultry contact patterns and address the first research question of my PhD thesis: *What is the frequency and extent of exposure to poultry in the general as well as occupationally exposed populations?*

3.1.1 Objectives of the Chapter

The objectives of this chapter are to:

- Describe the methods used in the cross-sectional survey of rural Cambodians which recruited and interviewed 3.600 rural Cambodians and 115 village chiefs from six provinces; and
- Describe the demographic characteristics and village characteristics of the recruited subjects.

3.2 Methods

3.2.1 Study Population

My first cross-sectional survey was carried out in six Cambodian provinces using a two-stage clustered sampling design (Bennett et al. 1991). All provinces and districts included in the study were identified for inclusion from a preliminary assessment of high poultry ownership and human population density (Figure 3-1) (NIS 1999; NIS 2002). Provinces located in the north-eastern (Steung Treang, Ratanakiri, Mondolkiri and Krecheh provinces), far eastern (Kach Kong, Palin) and south-eastern regions of Cambodia (Kach Kong and Kampong Som) were excluded from the study because they are largely mountainous, isolated and sparsely populated (Figure 3-1). Additionally, Kampot province, located in southern Cambodia and the location where 4 human H5N1 cases were confirmed, was not included in the study because prior to my involvement in Cambodia, almost all avian influenza related activities were focused in this province.



Figure 3-1 Population density in Cambodia

Map generated using data from a Columbia University population study done originally in 2005 and updated in 2006, and is a 2010 projection (pers. Comm.)

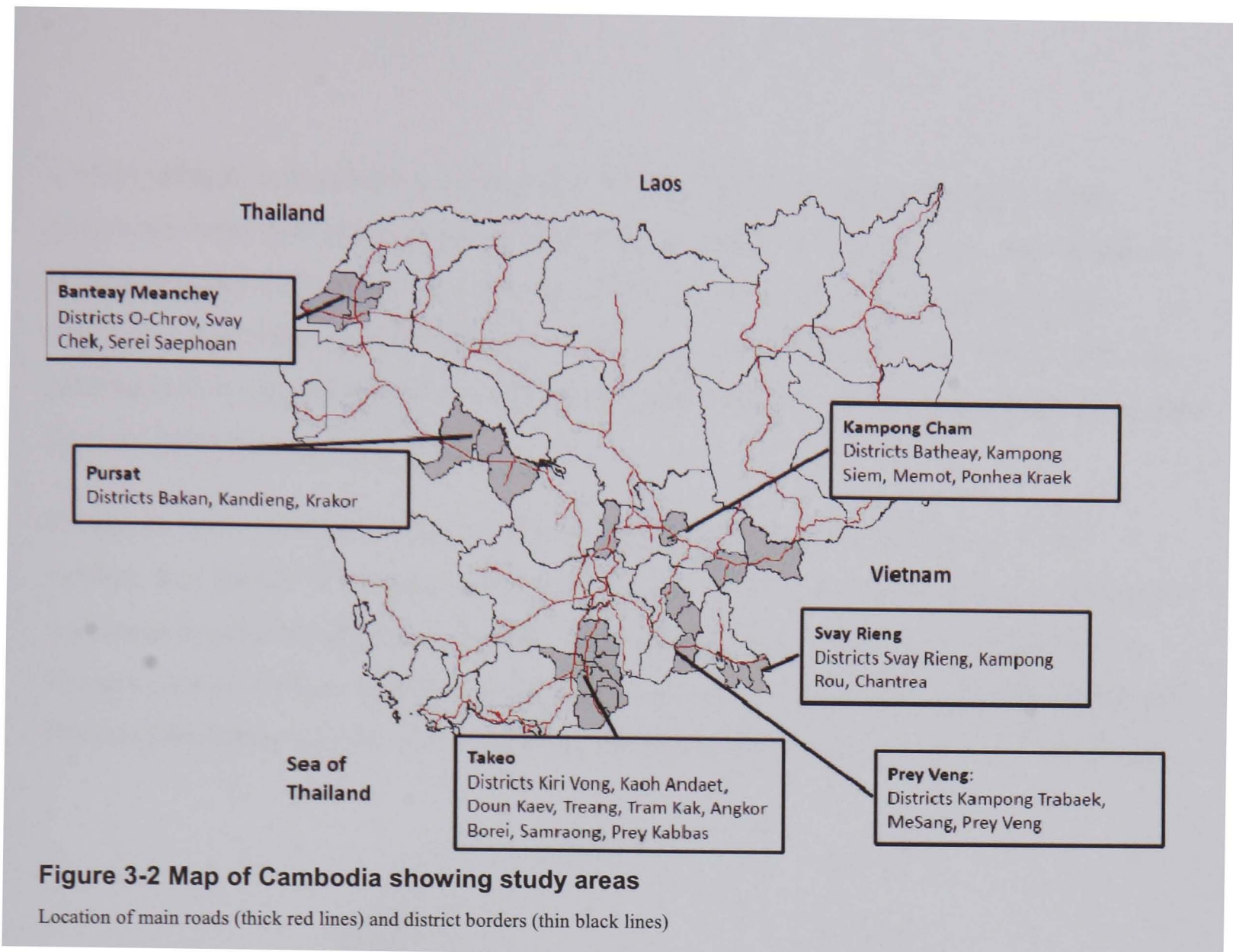
3.2.1.1 Description of the included study districts

Table 3-1 shows the number of villages and median population sizes in the included study districts. In addition to selecting districts with high poultry/human population density, districts were chosen for inclusion because of their potential cross-border trading activities with Thailand or Vietnam, or potential for wild bird mixing (Figure 3-2).

Table 3-1 Baseline population data of study areas

Province	Districts Included	Villages within included districts (n)	Mean village population size	Population range
Banteay Meanchey (BM)	3	195	761	105- 24,322
Kampong Cham (KC)	4	518	815	28 – 2,721
Prey Veng (PV)	3	378	755	134 – 2,882
Pursat (PR)	3	367	589	169 – 4,517
Svay Rieng (SV)	3	154	705	70 – 2,831
Takeo (TK)	8	907	621	81 – 4,724
Total	24	2519	708	70-24,322

Source (NIS 1999)



The districts in Pursat (Bakan, Kandieng, Krakor) were chosen for inclusion because of their proximity to the Tonle Sap Lake and the potential for wild bird mixing with domestic flocks. During the development phase of this study, discussions with members of NaVRI had indicated that it was possible that a large number of wild birds (e.g., wild ducks, turkeys) lived in the region of the lake. The western two-thirds of Pursat are largely mountainous and sparsely populated.

Banteay Meanchey is a province located in the north-west corner of Cambodia bordering Thailand. The districts in Banteay Meanchey (Chrov, Svay Chek, Serei Saephoan) were chosen for inclusion because of their potential cross-border trading activities with Thailand. The districts in Takeo (all), Prey Veng (Kampong Trabaek, MeSang, Prey Veng), Kampong Cham (Batheay, Kampong Siem, Memot, Ponhea Kraek) and Svay Rieng (Svay Rieng, Kampong Rou, Chantrea) provinces were selected because of their potential cross-border trading activities with Vietnam (Figure 3-2).

Twenty villages were included per province based on feasibility assessments (i.e., time constraints because of distance and road conditions, budget) using four interviewer teams. A random sample of 120 clusters (i.e., villages; 20 in each province) were selected using probability proportion to population size (PPS) methodology (Bennett et al. 1991). PPS was used because it was not feasible to conduct a study of a random sample of all rural Cambodians from all villages in each province.

Of special note is that at the time of data collection, H5N1 had not been suspected nor confirmed in poultry or humans in any of the villages in the study areas. However it had been confirmed in poultry and humans in one district where villages were randomly selected in Kampong Cham (Memot district) and one district in Prey Veng Province (Kampong Trabeak) (WHO 2006-2009). The locations of the randomly selected villages are shown in Figure 3-3.

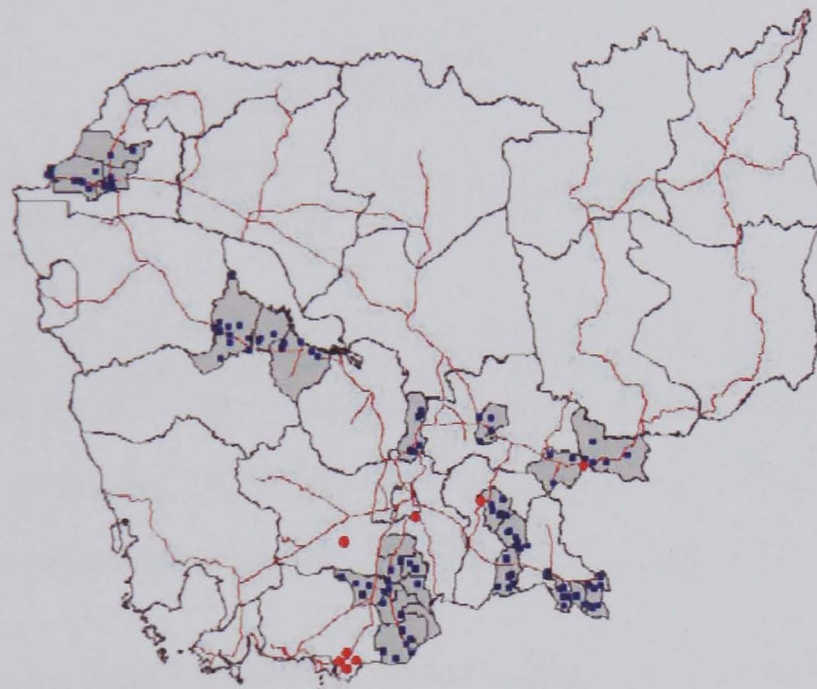


Figure 3-3 Map of Cambodia showing study districts (grey), the locations of the included villages (blue dots), and locations of the 8 human H5N1 cases

3.2.1.2 Sample size calculations

Sample size calculations were based on the precision with which an estimate of an exposure/behaviour (e.g., slaughter poultry) could be obtained assuming that the exposure/behaviour is 50% (resulting in a conservative estimate of the required sample size) (Figure 3-4). These calculations were applied separately to adult males (>15 years old), adult females and children

(≤15 years old¹) because these groups are of particular interest in terms of different poultry-handling practices resulting in potentially different exposures.

Figure 3-4 Sample size calculations

Formula to calculate the required sample size is shown below (EpiInfo v6.04)

$$n = \frac{DEFF \times N \times \hat{p} \times \hat{q}}{\frac{d^2}{Z_{1-\alpha/2}^2} \times (N - 1) + \hat{p} \times \hat{q}}$$

Where:

DEFF	Design effect	2
N	Population size	Estimated to be 1000000
\hat{p}	Estimated proportion in the target population with the event of interest	Estimated at 50% (conservative estimate)
$\hat{q}=1-\hat{p}$	1-0.5	0.5
d	Absolute precision	0.05
$Z_{1-\alpha/2}$	97.5 percentile of the standardized normal distribution	1.96

$$N = \frac{2 \times 1000000 \times 0.5 \times 0.5}{((0.05)^2 / 1.96^2) \times (1000000 - 1) + (0.5 \times 0.5)} = 768$$

Therefore, to estimate the behaviour with a precision of ±5 %, 6.4 (768 / 120 clusters) individuals in each group should be included from each village. This value was rounded up to 10 individuals per cluster to facilitate data collection.

Increasing the precision to ±1% (d=0.01) increased the sample size to 19,025/120 = 158.5 per cluster, which was not feasible. The village population sizes in the study areas precluded us from including 476 people (i.e., 159 adult males + 159 adult females + 159 children) per village.

The precision in my study using 3,600 subjects (10 adult females x 10 adult males x 10 children x 120 villages) is ±2.3%.

Therefore, to estimate the behaviour with a precision of ±2.3%, 10 individuals in each group (i.e., 30 individuals in total) were included from each village. Therefore the total number of subjects planned for inclusion in the study was 3,600 (30 subjects X 120 villages), in which 1,200 would be children ≤15 years old, 1,200 adult (>15 years old) males and 1,200 adult (>15 years old) females.

¹ Previous surveys conducted in Cambodia have classified adults as >15 years old. As such, children were defined as ≤15 years old.

A breakdown of included study provinces, districts, villages and quantity of study subjects is provided in Table 3-2. Data collection was conducted in two phases (1) November-December 2006 and (2) November- December 2007, which is the start of Cambodia’s dry season, to minimize problems of village accessibility from heavy rains. During the first phase of data collection (November-December 2006), four provinces were surveyed (Banteay Meanchey, Svay Rieng, Takeo and Pursat provinces). During the second phase of data collection (November-December 2007), two provinces (Kampong Cham and Prey Veng) were surveyed. Data collection for phase two took place exactly one year after phase one to minimize any seasonal differences in practices.

Table 3-2 Recruitment of rural Cambodians

Province	Districts	Villages (N)	Recruited Subjects	
			Village Chiefs	Subjects
Banteay Meanchey	O-Chrov, Svay Chek, Serei Saephoan	17 ^a	17	600 (200 adult males, 200 adult females, 200 children)
Kampong Cham	Batheay, Kampong Siem, Memot, Ponhea Kraek	19 ^a	19	600 (200 adult males, 200 adult females, 200 children)
Prey Veng	Kampong Trabaek, MeSang, Prey Veng	19 ^a	19	600 (200 adult males, 200 adult females, 200 children)
Pursat	Bakan, Kandieng, Krakor	20	20	600 (200 adult males, 200 adult females, 200 children)
Svay Rieng	Svay Rieng, Kampong Rou, Chantrea	20	20	600 (200 adult males, 200 adult females, 200 children)
Takeo	Kiri Vong, Kaoh Andaet, Doun Kaev, Treang, Tram Kak, Angkor Borei, Samraong, Prey Kabbas	20	20	600 (200 adult males, 200 adult females, 200 children)
Total 6	24	115	115	3,600 (1200 adult males, 1200 adult females, 1200 children)

^a Within Kampong Cham and Prey Veng, 1 village was selected by PPS twice because of its large population. In these villages 60 subjects and 1 village chief were interviewed; One village in Banteay Meanchey was selected 4 times using PPS and therefore 120 residents and 1 village chief were included from this village.

3.2.2 Questionnaire Development

Four separate standardized questionnaires were developed for the 1) village chief, 2) head of the household, 3) adult family members and 4) children. All questionnaires are provided in Appendix B. The aims of the questionnaires were to obtain information on poultry contact patterns and understanding of avian influenza at an individual level. All closed-ended questions were recorded as binary (yes/no) responses and frequencies of contact when evaluated were recorded as always, sometimes or never. All questions were precoded for ease of data entry.

3.2.2.1 Village chief

A one-page questionnaire was designed for the village chiefs to collect demographic information on the village (population, number of households) and the use of middlemen to trade poultry from their village (discussed further in Chapter 6).

3.2.2.2 Adult subjects

Subjects >15 years old were asked questions on the following topics:

Demographic Characteristics: Demographic variables included age (years old), gender, address (village, commune, district, province), occupation, the highest level of education completed (never attended school, primary, secondary, high school, higher), the country they were born in (Cambodia, Thailand and Vietnam) and religion. Each subject's ability to read and write was assessed by their competency to write and read a sentence in the presence of the interviewer. With the exception of occupation, all questions were closed-ended questions.

Poultry Contact: Subjects were asked if they had direct contact with domestic poultry through food preparation (slaughter poultry, remove or clean internal organs, cut or wash meat), caring for domestic poultry or fighting cocks (feed, clean animals or cages) and other activities (e.g., collect dead domestic/wild poultry for food, eat wild birds, remove feathers from sick poultry, attend fighting cock events). The nature of how Cambodians prepare poultry for consumption and care for poultry was evaluated by direct observation and informal questioning of adults living in rural Cambodia by myself in the field prior to piloting the questionnaires. The questionnaire also asked if they had indirect contact with poultry—as a proxy measure of exposure—in the immediate environment around the home and village via water sources (e.g., bathe/swim in ponds where poultry had access). All poultry contact questions and frequency of activity were asked as closed-ended questions.

A series of questions assessed awareness of avian influenza: source of AI information; knowledge of poultry-to-poultry, poultry-to-human, and human-to-human transmission; and knowledge of symptoms of AI in humans. Source of AI information and transmission questions were asked as closed-ended (yes/no) questions.

Poultry Mortality: Poultry mortality was evaluated as whether the household experienced poultry mortality since the Khmer New Year (closed-ended question; binary response) and the quantity of chickens and ducks raised, became sick and/or died from illness since the Khmer New Year. Through field visits, I learned that sickness in poultry is recognized by most villagers if their poultry suffer from seizures, have white eyes or become motionless. Subjects were asked if they reported poultry mortality, to whom they reported poultry mortality and the symptoms that poultry experienced that would warrant them to report poultry mortality. Poultry owners were asked about practices when poultry were sick and died from illness (e.g., prepare for household consumption, how were poultry carcasses disposed). They were also asked what they did with the poultry from a sick flock that remained healthy (e.g., prepare for household consumption, how were poultry carcasses disposed). With the exception of quantity questions, all poultry mortality questions were asked as closed-ended questions.

In addition, a series of knowledge and attitude questions about poultry mortality followed this section of the questionnaire and addressed whether subjects felt it was important to report poultry mortality and what would encourage and discourage poultry mortality reporting. Knowledge and attitude questions were asked as open-ended questions with some pre-recorded answers in the questionnaire. Pre-coded responses were several frequently stated responses to the knowledge and attitude questions identified during piloting of the questionnaires. We therefore included these responses as pre-recorded answers in the questionnaires with the addition of an answer of “Other, please specify” to minimize the amount of time to record any anticipated answers.

Basic Hygiene Questions: Basic hygiene questions including hand washing, household water source and the presence/absence of soap in the household were assessed via closed-ended questions and frequency was noted as always, sometimes or never.

Observation: At the end of each adult and head of household questionnaire, the interviewer was instructed to observe the household surroundings and fill in approximately 10 questions regarding chicken and duck ownership (e.g., do you see chickens/ducks in the house yard?) and if the flocks were not caged, the locations they were allowed to forage for food.

The adult questionnaire took approximately 30 minutes to administer.

3.2.2.3 Heads of household

In addition to all questions included in the adult questionnaire (as described above), the head of the household was asked questions about the following:

Household characteristics: Characteristics of the household, including the total number of people living in the house, total number of children ≤ 15 years old living in the house, house composition, and household asset ownership. With the exception of quantity questions, all household characteristic questions were asked as closed-ended questions.

Poultry and other animal ownership: The quantity of animals owned was collected differently for Phase 1 and Phase 2 of the data collection. During Phase 1, the quantity of animals owned by the household was collected as the total number of animals in the household at the time of the interview. During Phase 2, the quantity of animals owned by the household was collected as the total number of animals raised since the Khmer New Year (an 8-month time period [justification for this time period is described in Section 3.2.4]). Animal husbandry was assessed by asking and observing whether poultry flocks were free-ranging, mixed with other poultry and/or animals in the house yard and where flocks were allowed to forage for food (e.g., pond, rice fields). With the exception of quantity questions, animal husbandry questions were asked as closed-ended questions.

Fighting cock ownership: Fighting cock ownership, trading, morbidity and mortality were evaluated at the household level. The location(s) where fighting cocks were purchased and sold were asked as open-ended questions. Questions about the attendance at fighting cock events, where fighting cocks are kept in relation to the house, and frequency of fighting cock morbidity and mortality during the previous 8-month period were asked as closed-ended questions.

Poultry selling/trading practices (discussed further in Chapter 6): Poultry trading practices of the household were evaluated as the quantity and frequency of chickens sold to locations inside and outside of their village during the previous 8-month period. The use of middlemen for trading poultry was also assessed and the destination of the sale of poultry was recorded. The mechanism by which subjects transported poultry (e.g., truck, motorbike, bicycle) and the use of cages to transport poultry for trade was assessed. The destination and quantity of chicken and

duck sale was asked as an open-ended question. All other poultry trading practice questions were asked as closed-ended questions.

Wild bird mixing: Questions were included to address the extent of wild and domestic birds mixing. The head of the household was asked if their flock(s) mixed with wild birds, where they mixed (location around the house yard) and the species (if known) of the wild birds that mixed with their flocks.

Economic importance of poultry raising in the household: The head of the household was asked if their main source of income was from poultry raising, the total income generated from poultry raising per year and the affect on the household economy if they were to stop selling poultry. During Phase 2 of the data collection, I added a question to assess the proportion of households that vaccinated their flocks against H5N1.²

The head of household questionnaire took approximately 40-45 minutes to administer.

3.2.2.4 Children

A short questionnaire for children was developed to collect demographic information as well as basic direct and indirect domestic and wild bird poultry contact patterns. Interviewers were instructed to administer the questionnaire directly to the child. If they were unable to answer for themselves, the parent/guardian answered for them.

Poultry contact: Children were asked several questions regarding the care of domestic poultry and fighting cocks at their home; food preparation practices of domestic poultry; collecting, hunting and playing with domestic or wild birds, playing with sick or dead birds; and bathing in ponds which are accessed by poultry. All poultry contact questions were asked as closed-ended questions.

² Vaccination for H5N1 has not been implemented in Cambodia however I wanted to evaluate the perception of vaccination by poultry owners since there are non-government individuals in rural areas of Cambodia claiming to have bird flu vaccines for sale.

Knowledge of avian influenza: Seven questions about the child's understanding of AI were included in the questionnaire. Only children that were able to directly be interviewed without the assistance of a parent/guardian were asked knowledge of AI questions. With the exception of *have you heard of bird flu, can you get bird flu, and can you tell if poultry are sick*, all knowledge questions were asked as open-ended questions.

The child questionnaire took approximately 10 minutes to administer.

3.2.3 Questionnaire Translation

All questionnaires were translated into Khmer by Institut Pasteur of Cambodia (IPC) staff involved in the study and back translated into English by IPC staff unrelated to the study. Any discrepancies were evaluated by myself and corrected by Khmer IPC staff.

3.2.4 Recruitment of Staff, Interviewer Training and Questionnaire Piloting

Sixteen Cambodian interviewers (all university-level educated sociology students) were recruited and trained to administer the questionnaires in Khmer. A two-day training course was held prior to data collection, which included an introduction to avian influenza and objectives of the study; a thorough run through the questionnaires to ensure understanding of *each* question contained in the four questionnaires and piloting the questionnaires by the study teams. The two-day training focused on how to conduct structured interviews to minimize interviewer and information bias (Armstrong et al. 1992). The training manual provided to all interviewers is provided in Appendix C.

Questionnaire piloting was conducted first in Kampong Cham by myself and IPC staff prior to the two-day staff training and secondly in Kandal province by interviewer teams. In both instances, piloting was conducted to evaluate the content, wording, understanding, order and relevance of each question (Armstrong et al. 1992). During piloting of the questionnaires, we identified substantial difficulties with subjects recalling periods of time such as *In the past two months, have you ...?*. We found that subjects needed to be reminded of a major event in order to recall events and therefore, we piloted asking recall periods since a major Cambodian holiday and chose the Khmer New Year (mid-April annually) to refer the subjects to. When asking about an event that occurred in the past, subjects were asked to recall the event or

practice within the previous 8-months, i.e., between the time of the interview and the Khmer New Year Holiday period (mid-April annually). Therefore, in the finalized questionnaires, recall questions were asked as *Since the Khmer New Year, have you...?* Few other substantial changes were made to household surveys after piloting; however instructions (e.g., skip patterns) and questions (primarily to correct translation) were modified as needed.

3.2.5 Ethical Considerations

3.2.5.1 Informed consent

Two levels of consent were obtained for the survey. The first level of consent was for the village to participate in the study. Along with a representative from IPC and/or National Veterinary Research Institute (NaVRI), I met with the village chief to explain the purpose and methods of the study. Participation by the village was based on the independent decision of the chief. This level of consent is required before individuals living in villages are allowed to give individual informed consent to participate in a study.

The second level of consent was individual informed consent for participation in the study. Subjects or their parents/legal guardians (for subjects <18 years old) were informed in Khmer about purpose of the study, benefits and possible risks to the subjects, study procedures, voluntary participation and withdrawal, contact for answers to questions regarding the study, and confidentiality of subject records.

Informed consent was documented by the use of a written consent form approved by the Cambodian and LSHTM Ethics Committees and signed or fingerprinted³ and dated by the participant/parents, and by the trained interviewer (Appendix D). The signature/fingerprint confirms that the consent is based on information that has been understood. Each subject's signed informed consent form has been kept on file in a locked storage area at IPC and each subject or guardian received a copy of the information sheet.

³ In Cambodia, signing documents with a fingerprint is considered more rigorous than a signature. Cambodians believe that anyone can sign their name, but cannot reproduce their fingerprint.

3.2.5.2 Ethical approval

Ethical approval was granted from the Cambodian Ministry of Health and LSHTM. Copies of ethical approval are provided in Appendix E.

3.2.6 Data Collection

Prior to data collection, field visits were conducted by myself and a representative from IPC or the NaVRI in each of the 115 study villages. Meetings were held with provincial veterinarians and village chiefs to inform them of the study objectives and obtain verbal approval to conduct the interview in their districts and villages, respectively.

Four teams of five individuals (1 team leader, 3 interviewers and 1 local village guide) conducted structured interviews in Khmer with participants at their homes. At the start of each day, the team leader met with the village chief to draw a map of the village, identify village boundaries and identify a local guide to accompany the team around the village for the day. Within each village, the first household was chosen randomly from the centre of the village. Subsequent households were then systematically sampled using a sampling interval having been chosen at random by myself (using random number generation from 1 to 10) for each village until thirty people (i.e., 10 adult [>15 years old] males, 10 adult females and 10 children [≤ 15 years old]) plus 1 village chief were interviewed. Inclusion criteria for individuals included a) residence in village ≥ 6 months, b) minimum age of ≥ 1 year old and c) ability of adult subjects to hear and speak.

For the first five households visited in each village, team leaders were instructed to include one adult as the head of household. The remaining 15 adults included in the village were administered the adult questionnaire. I made no preference as to the gender of the head of household questionnaire as long as the 20 adults included in each village were composed of 10 males and 10 females. The team leaders were provided with a tally sheet to keep track of gender and subject type for each village (Figure 3-5).

Village Tally

Village _____

Household	Head of Household		Adult		Child	Interviews Completed per house
	Male	Female	Male	Female	Male/Female	
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						
Total						
Total Adult Male			Total Head Of Household			
Total Adult Female						
Total Child						

Figure 3-5 Team leader subject tally sheet used during field collection

Therefore for each village, among the 20 adult subjects required for inclusion, 15 were administered the adult questionnaire and five were administered the head of household questionnaire. As mentioned above the adult and head of household questionnaire were identical in all poultry handling behaviour questions with the exception that the head of household questionnaire included additional sections on household animal ownership, husbandry practices, fighting cock ownership and the economic importance of poultry raising in the household.

Interviewers were instructed to interview all household members that were present at the time of visit. For residents not at home at the time of initial visit, team leaders made an appointment to interview absent subjects later that day. Team leaders checked questionnaires for completeness and accuracy in recording by the interviewers at the end of each household visit. Any missing or unclear values on the questionnaire were corrected before the team left their allocated village each day. During each day of the data collection, I visited each study team in their allocated villages to monitor progress and address any questions by my field staff. At the end of each day, I checked the questionnaire data and discrepancies were corrected with the interviewers.

3.2.7 Constraints of Data Collection

Because of feasibility and safety issues (i.e., poor road conditions, distance of remote villages and increased potential for theft at night), each team had only one day (daylight hours approximately 6:30am – 5:30pm) to complete the 31 interviews in their allocated village. It was not possible to return to the villages the following day because of the distance of villages, road conditions, availability of interviewing staff and budget constraints. All villages were approximately 4 – 12 hrs away by 4X4 from Phnom Penh and 1-2 hrs from the district centres.

The interviewer teams did not have any problems completing the 31 interviews each day, although some teams needed to remain in the villages until late afternoon to complete interviews of returning subjects (mostly adult males) from work in the rice fields. It took one week to complete interviews of 600 subjects in each province (30 subjects x 20 villages) and therefore six weeks to complete interviews in all six provinces (4 interviewer teams x 30 subjects/village x 5 villages/week x 6 weeks = 3,600 interviews).

Figure 3-6 shows interviews that were conducted during the study. Verbal permission to take pictures and use them in presentations and/or reports was obtained from the interviewers, subjects and/or guardians.



Figure 3-6 Interviews of subjects recruited and included in the first cross-sectional survey of rural Cambodians

Verbal permission to take pictures and use them in presentations and/or reports was obtained from the interviewers, subjects and/or guardians.

3.2.8 Compensation

Each participant received an “avian influenza compensation kit” containing reusable rubber gloves and one bar of soap (worth approximately US\$1). Compensation kits were provided by UNICEF.

3.2.9 Data Entry

Two data entry analysts participated in the two-day training described in section 3.2.4 and were provided training on how to use EpiData v3.1 (EpiData association, Odense, Denmark) for data entry. The questionnaires were brought back to Phnom Penh where the two data analysts translated any open-ended questions recorded in Khmer into English and organized questionnaires by village and questionnaire type (head of household, adult male, adult female,

children, and village chief). All questionnaires were double entered into EpiData by the two data entry analysts and verified by checking discrepancies between the two data sets. Any discrepancies were checked against the original questionnaires and corrected. Additionally, the GPS codes of all surveyed households were entered into ArcGIS, version 9.0 (ESRI Systems, Redlands, CA, USA).

3.2.10 Statistical Methods

In general all numerical values were illustrated using frequency distribution and summarized by presenting the median and interquartile range (IQR) of the variable. All distributions were compared using non-parametric tests. Associations between categorical variables were tested using contingency table analysis.

3.2.10.1 Demographic and socioeconomic characteristics of rural Cambodians

The study population is described as the number of subjects recruited and interviewed for each subject type (head of household, adult subject, child subject, and village chief). Descriptive data on age is presented as median and IQR and is presented for both male and female subjects. Age distributions were compared using Wilcoxon rank sum tests for males and females separately. Gender is presented as the number and percent male for each subject type. Household composition and asset ownership are presented as numbers and percentages and compared across geographic regions. Associations between categorical variables were tested by cross-tabulation in a contingency table and chi-square tests or Fishers exact tests were used for statistical tests of significance as appropriate.

Socioeconomic characteristics (education level, ability to read and write, asset ownership, and house composition) are reported as number and percentages. Chi-square or Fisher's exact tests (as appropriate) were used to test whether these variables were differently distributed between age groups, for males and females separately.

3.2.10.2 Poultry contact behaviour

All poultry contact practices were evaluated as binary (yes/no) questions. Descriptive information in the form of numbers and percentages are presented on individual level poultry

contact behaviour. All analyses of poultry contact variables were stratified by age and gender. The distribution of food preparation variables by age among males and females is presented graphically. For categorical variables, chi-square tests were used to test whether these characteristics were differently distributed between age groups for males and females separately.

3.2.10.3 Knowledge and attitude questions

All knowledge and attitude questions that were addressed as closed-ended questions are reported as number and percentages by gender. Chi-square or Fisher's exact tests (as appropriate) were used to test whether these variables were differently distributed between age groups, for males and females separately.

3.3 Results

The demographic characteristics of the 115 village chiefs and 3,600 rural Cambodian subjects and the characteristics of the 115 randomly selected villages are presented below. Further results from the data collected by methods described in this chapter are presented in Chapter 4 (animal ownership, morbidity and mortality) and Chapter 5 (poultry contact patterns).

3.3.1 Subject and Living Characteristics

A total of 3,715 subjects were recruited and interviewed in the first cross-sectional survey, including 3,600 rural subjects (1,200 adult (≥ 15 years old) males, 1,200 adult females and 1,200 children) and 115 village chiefs (Table 3-3). There were no refusals from village chiefs and the refusal rate of rural Cambodians was low ($< 1\%$). Reasons for refusal included illness or that they were "too busy" to complete the interview. Refusals were replaced by further subjects.

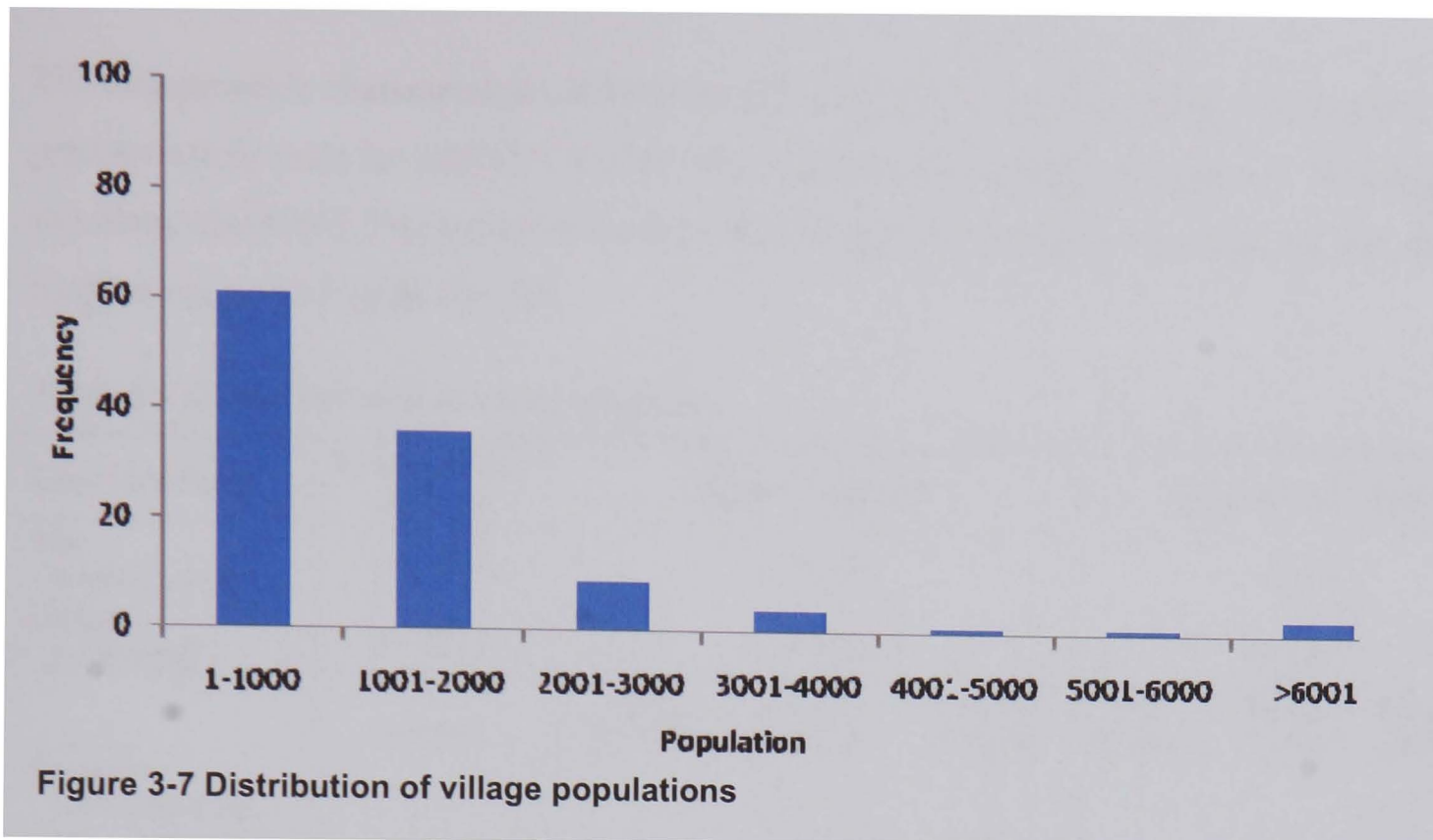
Table 3-3 Recruited subjects for the cross-sectional survey of rural Cambodians

Province	Districts	Villages (N)	Village Chiefs	Recruited Subjects		
				Poultry Owners	Questionnaires Used	
Pursat	Bakan, Kandieng, Krakor	20	20	600	{ 200 adult males 200 adult females 200 children	} 100 heads of household 300 adults
Banteay Meanchey	O-Chrov, Svay Chek, Serei Saephoan	17 ^a	17	600	{ 200 adult males 200 adult females 200 children	} 100 heads of household 300 adults
Svay Rieng	Svay Rieng, Kampong Rou, Chantrea	20	20	600	{ 200 adult males 200 adult females 200 children	} 100 heads of household 300 adults
Takeo	Kiri Vong, Kaoh Andaet, Doun Kaev, Treang, Tram Kak, Angkor Borei, Samraong, Prey Kabbas	20	20	600	{ 200 adult males 200 adult females 200 children	} 100 heads of household 300 adults
Kampong Cham	Batheay, Kampong Siem, Memot, Ponhea Kraek	19 ^a	19	600	{ 200 adult males 200 adult females 200 children	} 100 heads of household 300 adults
Prey Veng	Kampong Trabaek, MeSang, Prey Veng	19 ^a	19	600	{ 200 adult males 200 adult females 200 children	} 100 heads of household 300 adults
Total		115	115	3,600	1200 adult males 1200 adult females 1200 children	600 heads of household 1800 adults 1200 children

^a Within Kampong Cham and Prey Veng, 1 village was selected by PPS twice times because of its large population in the village selected twice 60 subjects and 1 village chief were interviewed; One village in Banteay Meanchey was selected 4 times using PPS and therefore 120 residents and 1 village were included from this village.

3.3.2 Village Demographics

Village characteristics were enumerated from the village chief questionnaires (n=115). The median population size of the villages included in the study was 951 (IQR: 643-1460, max 24,332; Figure 3-7). The median number of households in each village varied greatly (185; IQR: 120-294, max 7,000). The largest villages were located in Poipet, Banteay Meanchey (bordering Thailand).



The median distance from a village to the nearest health centre and hospital was 3.0 km (IQR: 2-5 km) and 11 km (IQR: 6-19 km), respectively. Roads between villages and health centres were most often made of dirt (74.8%); less common were paved roads (24.4%) and rivers (0.9%).

Household characteristics were enumerated from the head of household questionnaires (n=600). The median number of household members was 5 (IQR: 4-7, max 16). The median number of children ≤ 15 years old living in each house was 2 (IQR: 1-3, max 7). The majority of subjects lived in households that were built on piles above the ground (77.3%) and mainly composed of wood (70.2%); however 20.3% of subjects lived in households made of a combination of wood/straw, wood/cement (5.2%), cement only (4.3%), or mud/straw (3.0%) built on the ground.

The majority of households owned a bicycle (82.8%), approximately half owned a motorbike (45.8%), and few households owned a car (3.3%). Sixty-eight percent of households own a TV, and 52.8% own a radio. Ownership of bicycles ($X^2=20.2$, $p=0.001$), motorcycles ($X^2=33.1$, $p<0.001$), telephones ($X^2=33.2$, $p<0.001$) varied by province whereas ownership of cars ($X^2=7.9$, $p=0.16$), televisions ($X^2=9.2$, $p=0.10$) and radios ($X^2=10.0$, $p=0.8$) did not.

The demographic characteristics of subjects are shown in Table 3-4. Village chiefs were predominately male (n=108/115, 93.9%) and classified themselves as farmers⁴. The majority of village chiefs (67.5%) reported having only completed primary-level education and their median age was 52 (IQR: 48-58).

Table 3-4 Characteristics of study subjects

Characteristic	Village Chief (n=115)	Adults (n=2,400)			Children (n=1,200)		
		Total (n=2,399)†	Males (n=1,201)	Females (n=1,198)	Total (n=864) ^a	Males (n=445)	Females (n=419)
Age median (range)	52 (37-72)	36 (16-87)			9 (1-15)		
Gender n (% male)	108 (93.9)	1,201 (50)			612 (51)		
	n=114†						
Education n (%) reporting							
Never attended school	1 (0.9)	431 (18.0)	100 (8.3)	331 (27.6)			
Primary	77 (67.5)	1164 (48.5)	539 (44.9)	625 (52.2)			
Secondary	27 (23.7)	568 (23.7)	381 (31.7)	187 (15.6)	818 (94.7)	424 (95.3)	394 (94.0)
High school	7 (6.1)	185 (7.7)	144 (12.0)	41 (3.4)			
Higher	--	26 (1.1)	17 (1.4)	9 (0.8) ^b			
Pagoda	2 (1.8)	25 (1.0)	20 (1.7)	5 (4.2)			
Literacy ^c n (%) reporting		n=2,400	n=1,201	n=1,198	--	--	--
Can read	--	1,594 (66.4)	952 (79.2)	642 (53.5) ^b	--	--	--
Can write	--	1288 (53.7)	813 (67.8)	475 (39.6) ^b	--	--	--
Occupation n (% farmer)	n=115 97 (84.4)	1463 (61.8)	733 (61.0)	730 (60.9)	--	--	--

† Education level was not obtained from one female adult and one village chief

^a reported attending school (yes/no) among subjects ≥6 yrs old;

^b χ^2 p value <0.001 comparing adult males vs. adult females;

^c The ability to read and write was assessed by asking the subject to read and write a sentence for the interviewer, respectively (see Adult Questionnaire in Appendix B)

-- not evaluated

⁴ In Cambodia, the use of the word farmer for occupation is based on their own personal classification of their occupation, rather than the individual actually owning a working farm and raising animals or growing crops for profit in the commercial sector. In the majority of cases, subjects that classified themselves as “farmers” are actually rice farmers, rather than farmers raising animals. However I did not distinguish farmer type (e.g. rice, animal) in the questionnaire.

Among the rural Cambodians recruited in the study, half of the subjects were males (by study design), farmers (55%), predominantly Buddhist (99%) and born in Cambodia (99%). Age data were available for 3,598 of the 3,600 rural Cambodian subjects (99.9%). The age and gender of the household subjects and village chiefs is provided in Figure 3-8. The median age of the rural Cambodians in the study was 23.5 (IQR: 12-43). Among adult subjects, i.e., those older than 15 years of age, the median age was 36 (IQR: 24-49). The median age of adult men was 35 (IQR: 23-49; n=1,199), which was slightly less than the median age of adult women (37 [IQR: 25-49], n=1,199) (Wilcoxon p-value=0.04).

Among children, i.e., those less than 16 years of age, the median age was 9 (IQR: 5-12). The median age of boys was 9 (IQR: 5-12.5, n=612), which was not significantly different from the girls median age (8 [IQR: 5-12], n=588, Wilcoxon p-value=0.37).

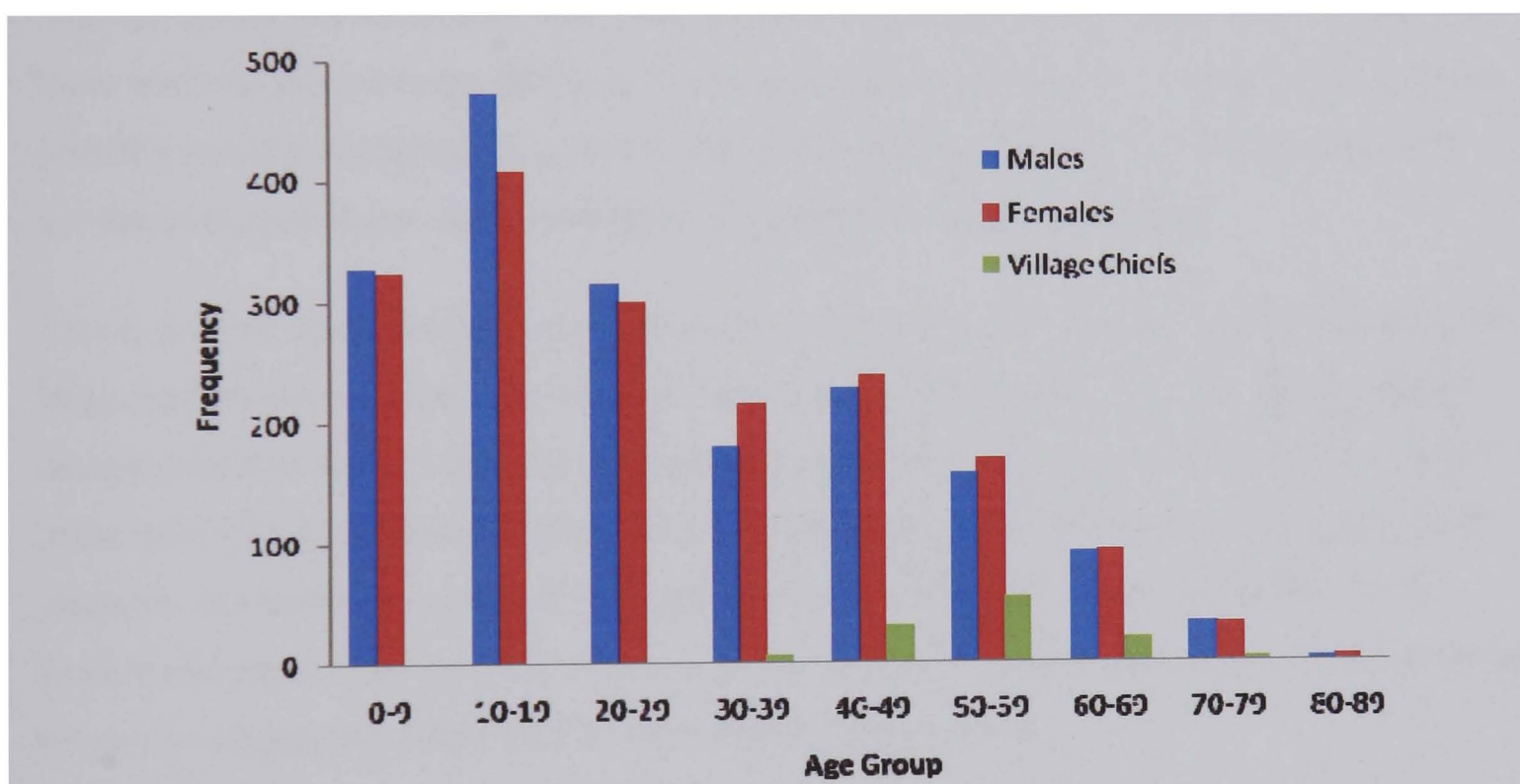


Figure 3-8 Age and gender breakdown of study subjects included in the survey (n=3,713)

Education level ($X^2=265.2$, $p<0.001$) and the ability to read ($X^2=178.0$, $p<0.001$) and write ($X^2=190.9$, $p<0.001$) were significantly higher among adult males compared to adult females and the ability to write was highest among adult respondents in Svay Rieng (63.0%) and lowest in Kampong Cham (41.5%, $p<0.001$). There were no differences by gender in education level among children ages 6-15 years old ($X^2=2.5$, $p=0.12$).

3.4 Discussion

This study is the first large-scale cross-sectional survey of a randomly selected population to evaluate the poultry handling behaviours of a population in regular contact with poultry and is the first study to evaluate poultry contact patterns of children. Direct contact with infected birds is assumed to be the main source of infection to humans (Writing Committee of the Second World Health Organization Consultation on Clinical Aspects of Human Infection with Avian Influenza 2008) although the specific mode of effective transmission from animal-to-human is not fully understood and the absence of detailed exposure data from the approximately 400 human H5N1 cases makes it impossible to ascertain the true risk factors for human H5N1 cases. To fully evaluate risk factors for H5N1 infection, a large-scale seroprevalence study testing for antibodies of H5N1 would have been ideal; however at the time this thesis was developed, the funds available to conduct such a study were limited and there was reluctance among ethics review boards to grant approval for a study that could have possibly resulted identifying few, if any, seropositive individuals as had been the case in a previous seroprevalence study conducted in Cambodia (Vong et al. 2006).

This is the first study that has been conducted to evaluate detailed poultry contact patterns of a large random sample of people living in daily contact with poultry. With the questionnaires designed for this study, I was able to capture detailed information on individual level contact patterns of a large random sample of rural Cambodians. As will be shown in the subsequent chapters, this study has captured information on how individuals come into contact with poultry and gender and age differences in poultry contact patterns. The results of this study can be used to inform any future HPAI/H5N1 seroprevalence studies.

This study has combined self-reported and observational data collection, particularly on questions regarding raising poultry. A short observation based questionnaire should accompany questionnaires that evaluate animal ownership and husbandry practices. Although, I found no differences in the proportion of households that self-reported owning chickens or ducks or if domestic flocks were free-ranging, there were difficulties in households reporting ownership of fighting cocks since fighting cocks are often raised by other individuals who are hired to care for the birds but do not actually own the birds themselves. Households that raised

and cared for fighting cocks reported owning no fighting cocks and thus the quantity of animals owned was reported as the number of fighting cocks observed in the household.

There are several limitations inherent to any studies with a cross-sectional design. Although we included a random selection of individuals from 24 districts in six provinces and Phnom Penh, we did not include subjects from Cambodia's 17 additional provinces. In particular, we did not include the indigenous tribes that reside in Cambodia's North-Eastern provinces, who may have different customs and rituals resulting in different contact patterns with poultry.

Therefore I cannot assert that the results of this study are representative of all Cambodia, but they are likely to be representative of the provinces initially identified as having high poultry ownership and human population density and hence typical of populations with high backyard poultry ownership (for their own consumption).

Secondly, the accuracy of the responses of children needs to be treated with caution.

Interviewers were instructed to administer the questionnaire directly to the child and although I monitored the technique of each interviewer who was responsible for interviewing children and was satisfied with their technique, I was not able to witness every child interview. Children were asked about their daily activities as they relate to contact with domestic and wild poultry and although I did not ask them to recall practices over any past period of time, the accuracy of 74% of children that answered questions directly may vary due to intraindividual variability in recall of regular activities (e.g., (Stein et al. 1991)). Conversely, the accuracy of the child's reported practices by a surrogate (i.e., parent or guardian) may vary as well (e.g., (Daly et al. 1994)).

After analyzing the data and having participated in several IPC H5N1 activities during my time in Cambodia, including a human and poultry outbreak investigation, two human seroprevalence studies and a case-control study to identify risk factors of H5N1 infection (Vong et al. 2009), I realized that several additional questions should have been included in the questionnaire. These include:

- The collection of quantity of cats owned by the household.

- The collection of more specific information on the grazing patterns of domestic flocks outside of the village i.e., the location where and frequency in which domestic flocks, specifically ducks, forage for food. Without this, I was only able to state whether flocks were free-ranging or not and if they were allowed to graze in rice fields. I did not, however, address where those rice fields were or the type of rice fields in which domestic ducks graze (rain fed low- or uplands, areas of deepwater/floating cultivation, or dry season irrigated land) (FAO 2004b).
- The collection of farmer type for self-reported occupation. The self-reported description of occupation should have included the type of farmer rather than just farmer. Without this information, it is impossible to distinguish between animal farmers vs. rice farmers. The predominant occupation in rural Cambodia is rice farming (FAO 2004b).
- The collection of household income. I intentionally did not collect household income because during piloting of the questionnaire we found that this was a sensitive issue and that subjects were reluctant to provide this information. Therefore, this variable was intentionally excluded from the head of household questionnaire. However this would have been an obvious indicator of socioeconomic status.
- The extent and frequency of swimming, bathing and fishing in ponds by adult subjects was not captured for all adult subjects included in this study. It was added into the questionnaires used in Phase 2 of the data collection and therefore is only available for 800 of the 2,400 adults included in the study.
- The collection of data on the type of farm equipment used, the proportion of households that share farm equipment and the frequency and locations in which the farm equipment is shared. Within Cambodia, the use of farm equipment is virtually nonexistent however, when it is used during rice harvesting season, it is often shared to minimize costs among rice farmers. Spreading of HPAI could be facilitated by the sharing of farm equipment (FAO 2005). The extent of the use and sharing of farm equipment should be further explored to adequately determine the potential risk of transmission of HPAI from farm-to-farm.

- Criteria used by FAO to define poultry production sectors (FAO 2006). Although I collected some data on the following of FAO's criteria: biosecurity, birds kept, shed, contact with other chickens, contact with other ducks, contact with other domestic birds, contact with wild life and some information on market outputs, I did not collect data on the following criteria: veterinary service, source of medicine and vaccine, source of technical information, breed of poultry, and food security of owner. These data would have allowed me to fully describe the poultry sectors in rural Cambodia according to FAO.

Results from this study are presented in the following two chapters. Chapter 4 presents data on animal ownership and husbandry, poultry mortality experienced and poultry mortality reporting of rural Cambodians. The chapter also explores poultry ownership in rural Cambodia using poultry sector definitions from FAO (FAO 2006) and proposes newly defined categories within Sector 4 holdings to describe the heterogeneity in husbandry practices within this sector.

Chapter 5 describes the extent and frequency of poultry handling behaviours of subjects and how they differ by age and gender. Using risk assessment methods, patterns of contact with poultry were used to generate risk indices of potential H5N1 transmission to different populations in contact with poultry.

Chapter 4 Results 1 of the Cross-Sectional Survey of Rural Cambodians: Animal Ownership and Husbandry, Poultry Mortality and Understanding of Avian Influenza

4.1 Introduction

Millions of people around the world, particularly in Asia and many parts of Africa, live in close proximity to domestic poultry (Epprecht & Robinson 2007; Gilbert et al. 2007). Although ownership of domestic poultry is believed to be common, flock size is usually small and primarily raised for household consumption (Burgos et al. 2008; Omiti & Okuthe; Sumiarto & Arifin 2008). Such flocks commonly mix with other domestic species such as pigs, cattle, dogs and cats increasing the possibility of reassortment of H5N1.

The Food and Agriculture Organization (FAO) has classified poultry production systems into four sectors (Table 2-4) in which they describe Sector 1 as the poultry production system for industrial integrated production; Sectors 2 and 3 describe commercial poultry production with decreasing levels of biosecurity, respectively, and Sector 4 describes village or backyard poultry production (FAO 2006). These definitions are most applicable in describing the poultry sectors of countries involved in varying levels of commercial poultry production.

They may not, however, be adequate to distinguish between the heterogeneous practices of backyard poultry holdings within countries throughout SE Asia and Africa, areas which in which HPAI/H5N1 has spread in domestic poultry populations (FAO 2008). Although it has been suggested that backyard poultry may be responsible for the sustained transmission of H5N1 within domestic and to/from wild-bird populations (e.g., (Gilbert et al. 2006; Grain 2006)), detailed information regarding the husbandry practices of countries with predominant backyard poultry holdings (FAO Sector 4) has not been provided. Without understanding the diversity of husbandry practices of Sector 4 farms, control policies that are typically developed for the average farmer may be inappropriate for a large majority of households that keep poultry (Chambers et al. 1993; Dent et al. 1994; Dent & Thornton 1998).

In this chapter, I describe the husbandry practices of households raising poultry and propose new subgroups for Sector 4 (village or backyard poultry) within the context of the current FAO sector definitions using data collected from my first cross-sectional survey of rural Cambodians. Data on poultry mortality occurring in the study areas are also presented as well as poultry mortality reporting practices.

4.1.1 Objectives of the Chapter

The objectives of this chapter are to:

- Describe poultry ownership and husbandry practices of study subjects in the context of FAO's poultry production sector definitions;
- Describe poultry mortality experienced and poultry mortality reporting practices of poultry owners;
- Describe the understanding of avian influenza of study subjects by age and gender; and
- Discuss the extent to which the rural poultry sector may have influenced HPAI circulation in Cambodia

4.2 Methods

The methods used to collect data used in this chapter have been presented in Chapter 3. Statistical analyses used in this chapter are provided below.

4.2.1 Animal Ownership and Husbandry Practices

Descriptive statistics were obtained for quantitative and qualitative variables related to animal ownership and husbandry practices. Keeping poultry fenced and separated from other domestic and wild animals was used as an indicator of biosecure husbandry. After evaluating the distributions of household chicken and duck ownership, chicken flocks were divided into four size categories (none, 1-10, 11-50 and >50 animals) and duck flocks into five categories (none, 1-10, 11-100, 101-1,000 and >1000 animals).

The hypothesis that flock size differs between geographic areas was tested by Kruskal-Wallis test. The hypotheses that the proportion of biosecure farms and the proportion of flocks mixing with wildbirds differs between geographic areas was tested by Chi-square and Fisher's exact tests (as appropriate).

4.2.2 Poultry Production Systems in Cambodia

Poultry production in rural Cambodia was classified under FAO's definitions for poultry production sectors (Table 2-4) using data from the 600 heads of household. In the questionnaires, I collected data on the following FAO criteria: "birds kept", "shed", "contact with other chickens", "contact with other ducks", "contact with other domestic birds", "contact with wild life" and some information on "market outputs" (described further in Chapter 6). According to the FAO classification system, "biosecurity" is listed as "high", "moderate-high" or "low" although definitions of these levels of biosecurity are not indicated. In their classification of poultry sectors, FAO includes criteria that are clearly part of the concept of biosecurity (shed, contact with other chickens, contact with other ducks, contact with other domestic birds, and contact with wild life); indicating that these criteria overlap with the term "biosecurity."

I collected information on what I believe are the key parameters that define biosecurity, i.e., how birds are kept (always indoors, free ranging) and contact with other animals (domestic and wild birds). I did not collect data on the following criteria: "veterinary service", "source of medicine and vaccine", "source of technical information", "breed of poultry" (other than chickens or ducks), and "food security of owner". Definitions of these criteria have not been provided in any FAO reports.

4.2.2.1 *Evaluating the heterogeneity in Cambodia's Sector 4 holdings*

To explore and evaluate the heterogeneity of Sector 4 holdings in Cambodia, new criteria are proposed to describe backyard flocks of subjects recruited in this study. Table 4-1 provides definitions of proposed subcategories within Sector 4 holdings (sub-categories A, B, C and D) using FAO defined variables (FAO poultry sector, description of poultry production) some modified FAO criteria (where birds are kept and contact with other domestic and wild animals) and newly proposed criteria (predominant species raised, flock size, and selling characteristics). FAO uses "market outputs" as a criterion but this is not well defined.

Table 4-1 Proposed subcategories within sector 4 poultry production

Criteria	Proposed sub-categories				
	Sub-category A	Sub-category B	Sub-category C	Sub-category D	
FAO Defined	FAO Poultry Sector	4	4	4	4
	Poultry Production	Village/ backyard	Village/ backyard	Village/ backyard	Village/ backyard
Biosecurity Measures	Biosecurity ^a	Minimal	None	Minimal	None
	Contact with other animals				
	Domestic poultry	Some	Yes	Some	Yes
	Wild birds	Some	Some	Some	Some
	Other domestic animals	Some	Yes	Some	Yes
	Birds Kept	Indoors Closed System or; Kept enclosed part of the day	Outdoors Open - Free-ranging	Indoors Closed System or Kept enclosed part of the day	Outdoors Open - Free-ranging
Flock Raising Characteristics	Predominant Species Owned [†]	Duck	Duck	Chicken	Chicken
	Flock Size [†] (median, range)	Med/Large >50	Small 1-50	Med/Large >50	Small 1-50
	Selling Characteristics				
	Sell eggs	Some	No or rarely	Some	No or rarely
	Sell birds ^{‡‡}	Some	No or rarely	Some	No or rarely

^aBiosecurity is newly defined as minimal and none using the criteria “contact with other animals” and where “birds are kept.” The cells shaded in yellow are modified FAO criteria; the cells shaded in red are newly proposed criteria.

[†]Flock Size: Small= 1-50 animals, Medium = 500-1,000, Large = 1001-10,000

[‡] For households that owned both chickens and ducks, the quantity of birds owned determined whether the household reared predominantly ducks (Subcategory A, B) or chickens (Subcategory C, D).

^{‡‡} Selling birds outside of the home village, (i.e., birds entering live poultry markets)

4.2.2.2 Definitions of the proposed sub-categories

Subcategory A are medium sized duck flocks (>50 birds) with minimal bio-security (birds are kept indoors or allowed to be free-ranging for part of the day/kept indoors or fenced in at night) and some input into the poultry market chain (poultry may be sold outside of their home village); Sector 4 Subcategory B are small duck flocks (≤50 birds) with no biosecurity (all birds are free-ranging and may mix with other domestic and wild animals) and minimal input into the poultry market chain (i.e., rarely sell poultry); Sector 4 Subcategory C are medium sized chicken flocks (>50 birds) with minimal biosecurity and some input into the poultry

market chain; and Sector 4 Subcategory D are small chicken flocks (≤ 50 birds) with no biosecurity and no input into the poultry market chain.

4.2.3 Poultry Morbidity and Mortality

For each of the studied flocks, the proportion of birds that became ill and the proportion of birds that died within the flock during the previous 8-month period (*Since the Khmer New Year...*)⁵ were calculated to obtain an estimate of background patterns of morbidity and mortality experienced when an outbreak of HPAI/H5N1 is not suspected or reported. Questionnaires used for adult and head of household subjects were edited for Phase 2 of the data collection (Kampong Cham and Prey Veng, Nov-Dec 2007) to more accurately obtain the necessary data. Illness among chickens and ducks was defined as illness perceived by the subject.⁶ The proportion of the flock that reportedly became ill or died during the previous 8-month period were calculated as:

Proportion of flock that became ill since the Khmer New Year:

$$= \frac{\text{Total \# of birds that became ill since the Khmer New Year}}{\text{Total birds owned since the Khmer New Year}}$$

Proportion of flock that died since the Khmer New Year:

$$= \frac{\text{Total \# of birds that died since the Khmer New Year}}{\text{Total birds owned since the Khmer New Year}}$$

Within-flock morbidity and mortality are numerically and graphically summarized and the hypothesis that they differ between geographic areas was assessed using chi-square or Fisher's exact tests, as appropriate. The proportion of households reporting poultry mortality were evaluated by within-flock mortality rates and was also evaluated after categorizing households as experiencing $>60\%$ mortality in a chicken flock or $>30\%$ mortality in a duck flock.

⁵ As described in Section 3.2, the questionnaires used in phase two of the data collection (Nov-Dec 2007) were modified to collect information on the quantity of chickens, hens and ducks owned, became ill and died from illness within an 8 month period of time. Therefore, morbidity and mortality rates were calculated for these two provinces (Kampong Cham and Prey Veng) only.

⁶ During field visits and piloting, we learned that illness among chickens and ducks is recognized by most villagers if their poultry suffer from seizures, have white eyes or become motionless.

To whom subjects reported poultry mortality was evaluated between geographic areas using chi-square or Fisher's exact tests, as appropriate. Reasons for reporting poultry mortality were evaluated by geographic region using chi-square or Fisher's exact tests, as appropriate.

4.2.4 Understanding of AI

All knowledge and attitude questions that related to the subjects understanding of how AI affected their flocks—including the attitudes about poultry mortality reporting, practices of poultry owners when poultry were ill or died, and knowledge of AI symptoms in flocks—that were addressed as closed-ended questions summarized as absolute and relative frequency of responses, by gender. Chi-square or Fisher's exact tests (as appropriate) were used to test whether these variables were differently distributed between age groups, for males and females separately.

4.2.5 Economic Importance of Poultry Rearing in the Household

The proportion of households that report to base their income on poultry raising and the annual income reported to be based on poultry raising is summarized as the median and range.

4.3 Results

4.3.1 Animal Ownership, Husbandry and Home/Farm Biosecurity

Most households included in the study owned chickens (83.7%) or ducks (35.7%) (33.2% owned both chickens and ducks), however most poultry flocks were small (Table 4-2).

Prevalence of other bird ownership (geese, singing birds, fighting cocks) was low, while most households owned pigs (55%), cattle/water buffalo (63.5%) and dogs (75.5%).

Table 4-2 Animal ownership among households in rural Cambodia (n=600)

Animal*	Households raising (%)	Median flock size (n)	IQR	Max
Chickens [†]	502 (83.7)	14	7-25	150
Ducks [‡]	214 (35.7)	7	3-15	1,900
Fighting cocks	23 (3.8)	2	1-5	30
Geese	3 (0.5)	3	2-6	6
Singing birds	20 (3.3)	1	1-1	6
Pigs	330 (55.0)	2	1-4	40
Dogs	453 (75.5)	2	1-3	12
Cattle/buffalo	381 (63.5)	3	2-5	20
Chickens and ducks	199 (33.2)	--	--	--
Chicken or ducks and pigs	323 (53.8)	--	--	--

*Ownership of cats was not assessed

[†]Most chickens are local breeds (Sampov, Moan Prey, Red Jungle fowl, Kragnas)

[‡]Most ducks are local breeds (Muscovy, Tear Angkam [layer], Tear Sampov [layer/meat])

Chicken and duck flock size varied by geographic region with the largest size chicken flocks located in Prey Veng Province (median flock size=20; Kruskal-Wallis $p<0.001$) and the largest duck flocks were located in Takeo Province (median flock size=12; Kruskal-Wallis $p<0.001$) (Figure 4-1).

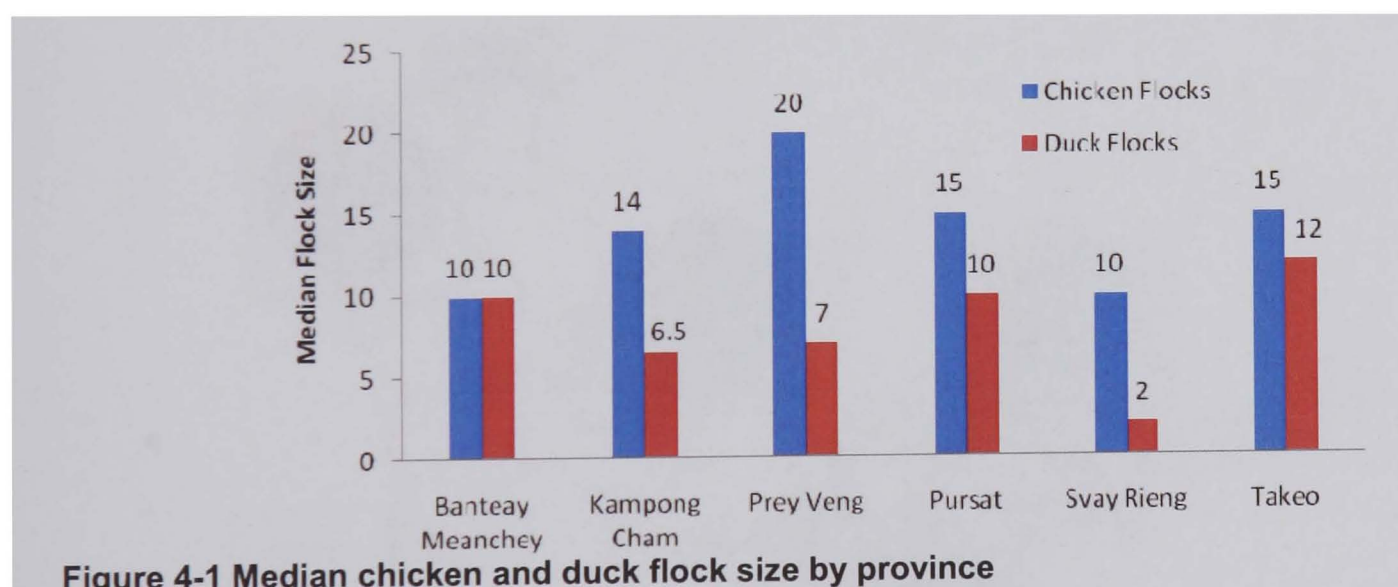


Figure 4-1 Median chicken and duck flock size by province

The distributions of chicken and duck flock sizes using flock size groupings are shown by province in Figure 4-2. The largest chicken flocks were located in the southern provinces of Svay Rieng and Takeo Provinces (maximum flock size=150) and the largest duck flocks (maximum flock size=1,900) were located in Takeo and Pursat provinces.

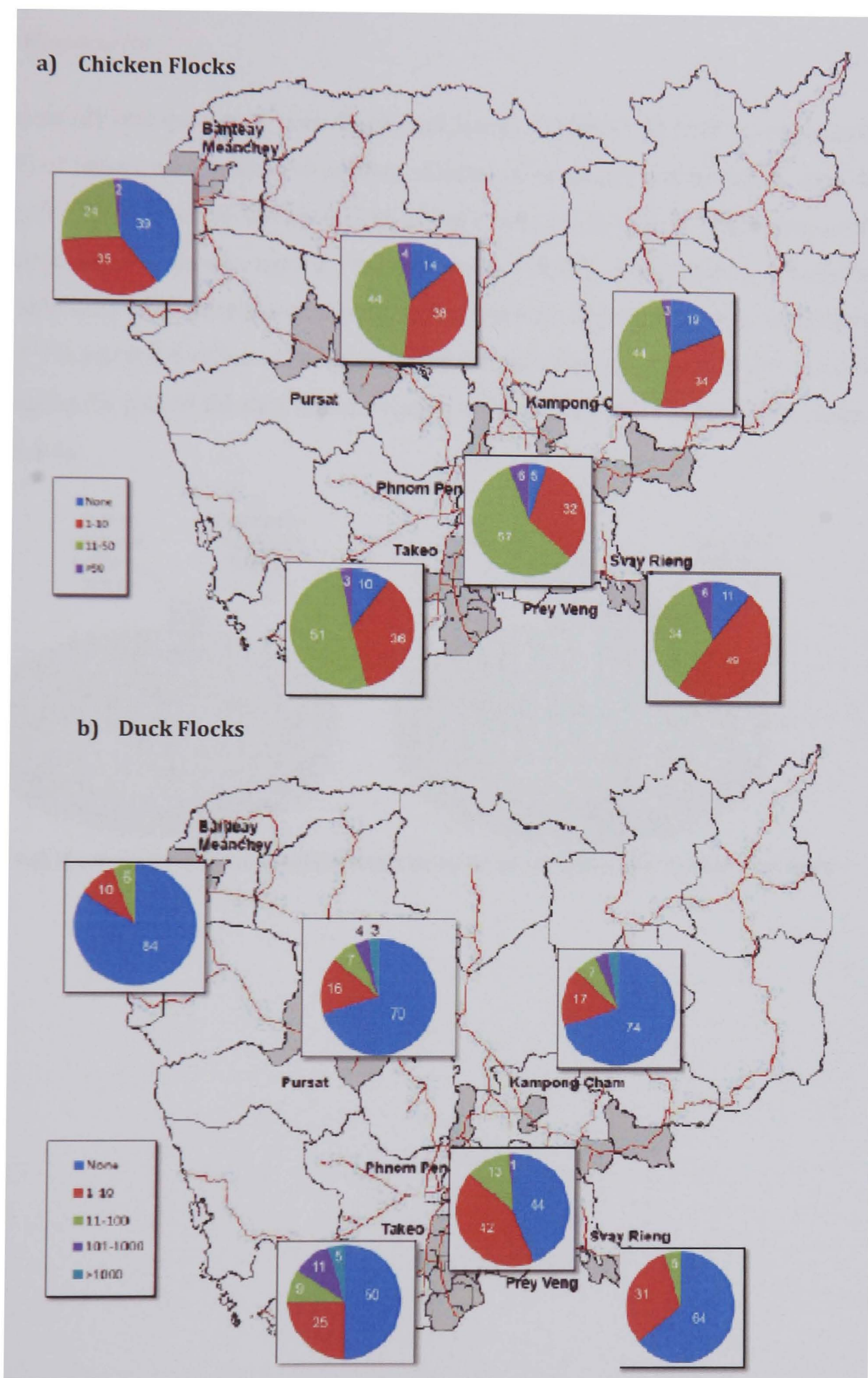


Figure 4-2 Chicken (a) and duck (b) flock size by province

4.3.1.1 Biosecurity

Approximately one quarter of households had fences (135/600, 22.5%), however only 8.9% (12/135) of fences were sealant to poultry. Almost all domestic poultry flocks were free ranging (97.8% of chicken flocks; 90.7% of duck flocks, Figure 4-3) and mixing with pigs and other domestic animals was reported to be common (53.8% of households owned poultry and pigs, Table 4-2). The proportion of households that kept birds enclosed in buildings at all times (4.7% and 0.8% of household raising ducks and chickens, respectively) or allowed to be free-ranging for part of the day/kept indoors at night was low (4.7% and 3.3%, respectively) (Figure 4-4).

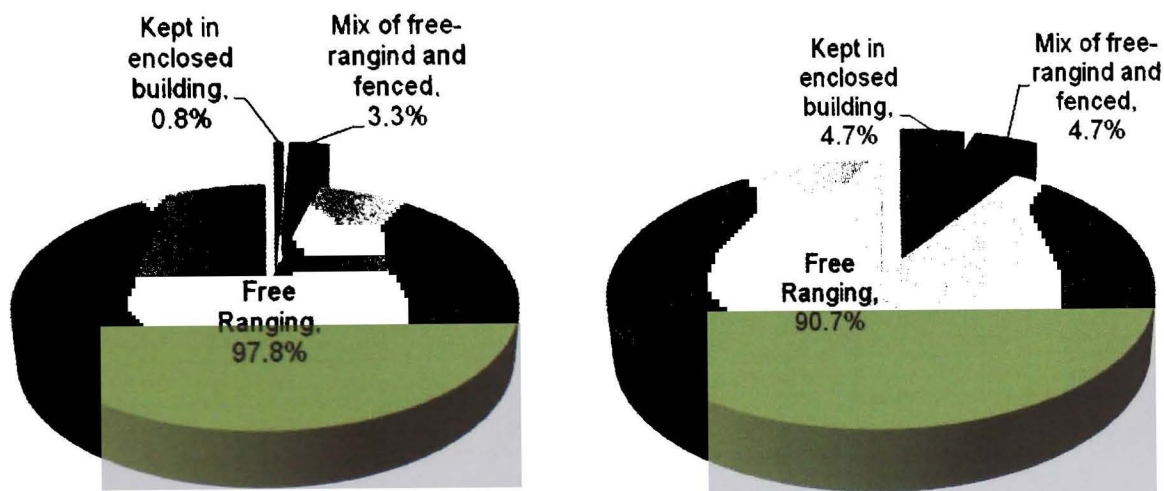


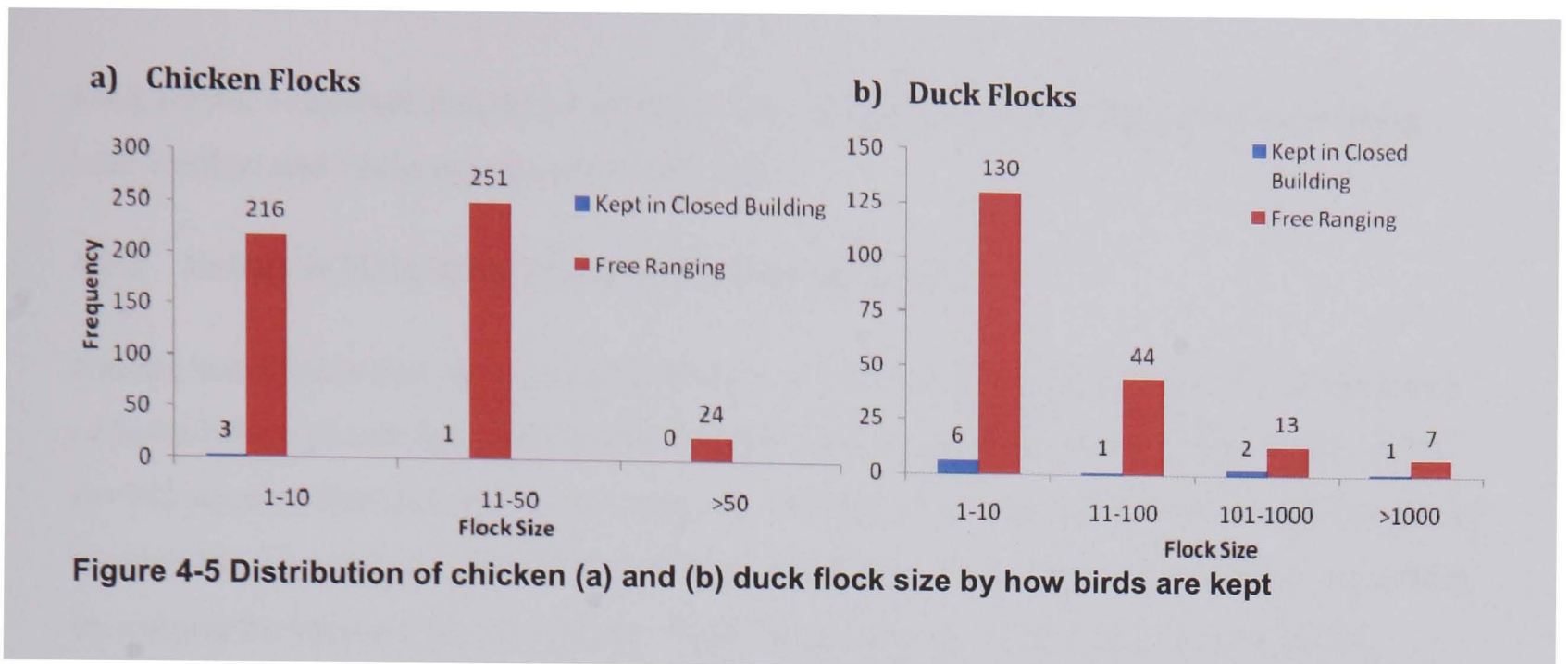
Figure 4-3 Husbandry characteristics used in chicken (left) and duck (right) raising



Figure 4-4 Husbandry practices of duck flocks

The top two photographs provide examples of free-ranging flocks and the bottom two examples of duck flocks and the use of fencing to restrict the area where ducks forage for food; however the water sources where these ducks roam are shared spaces with other flocks.

The proportion of households keeping free ranging chickens or ducks did not differ by geographic region (chicken flocks $\chi^2=8.0$ $p=0.16$; duck flocks $\chi^2=8.5$ $p=0.13$). Free-ranging poultry are allowed to forage for food in and around the house yard and in nearby rice fields but did not generally roam outside of their home village (observation only). Less than one percent (0.8%) of chicken flocks and 4.7% of duck flocks were always kept contained. The distribution of flock size stratified by where birds are kept (flocks that are free ranging vs. always kept enclosed) are shown in Figure 4-5.



4.3.1.2 Domestic poultry access to household water supply

Within the study areas, the primary water source of households is located outside of the home and includes water wells with a pump (46.3%), ponds (26.5%), open water wells (27.0%), lakes (2.5%) and rivers (3.3%). Poultry have access to many of these water sources, including ponds (44.6%; Table 4-3).

Table 4-3 Household water source and poultry access

Water Source	Primary water source of households n (%)	Water Source with poultry access n (%)
Pond	159 (26.5)	70/157 (44.6)
Lake	15 (2.5)	5/15 (33.3)
River	20 (3.3)	18/20 (90.0)
Open water well	162 (27.0)	--
Water well with pump	278 (46.3)	--
Water tap	35 (5.8)	--

-- no access possible

4.3.1.3 Vaccinated flocks

As described in Chapter 3, vaccination status of domestic poultry flocks was evaluated from households in Kampong Cham and Prey Veng only. Among these households (n=200), 3% of the heads of the household reported that they had vaccinated poultry flocks against AI. Among

those (n=6), 3 reported that they vaccinated their chicken flocks and 3 reported vaccinating both chicken and ducks against avian influenza.⁷

4.3.2 Extent of Wild Bird Mixing with Domestic Flocks

Among households that raise poultry (chickens or ducks, n=517), 25.3% (n=131) of the heads of household reported that their domestic flocks *sometimes* mix with wild birds, while 14.7% (n=76) reported that their flocks *always* (i.e., everyday) mix with wild birds. The proportion of households reporting domestic and wild bird populations (*sometimes* or *always*) mixing varied by geographic region with the highest proportion reported in Svay Rieng (65.2% 58/89), followed by Banteay Meanchey (56.5% 35/62), Takeo (54.1% 53/98), Pursat (50.6% 44/87), Kampong Cham (13.3% 11/83) and Prey Veng (6.1% 6/98) ($X^2=109.1$ p<0.001).

The wild bird species that were most commonly reported that mix with domestic flocks were “singing birds” (49.8%, 102/205) and egrets (19.5% 40/205). The locations where domestic flocks mix with wild birds include lakes (1.5%, 3/206), rice fields (60.7%, 125/206), and in the household farm yard (38.8% 80/206).

4.3.3 Overview of Cambodia’s Poultry Sector in Rural Cambodia

Using data from the 600 heads of household and FAO’s definitions for poultry production systems (**Error! Reference source not found.**), 98.3% of poultry flocks fall within Sector 4 holdings and the remaining 1.7% of poultry flocks fall within Sector 3 holdings.

The same data on poultry ownership at the household level is categorized using the proposed sub-category Sector 4 definitions provided in Table 4-1. Using the proposed definitions, poultry raising in the study areas can be described as 1.7% Sector 3 (semi-commercial), 0.7% Sector 4 Subcategory A (medium sized duck flocks with minimal bio-security; some input into the poultry market chain), 6.3% Sector 4 Subcategory B (small duck flocks with no biosecurity, no input into the poultry market chain), 0.2% Sector 4 Subcategory C (medium sized chicken flocks with minimal bio-security; some input into the poultry market chain), and 91.2% Sector

⁷ Note that there are currently no authorized vaccines for H5N1 available in Cambodia. However there may be non-government individuals that claim to have “vaccines” for poultry diseases.

4 Subcategory D (small chicken flocks with no biosecurity, no input into the poultry market chain) indicating that the majority of households involved in raising poultry in rural Cambodia keep a very small number of free-ranging chickens.

4.3.4 Poultry Mortality and Reporting

Among households that owned poultry (n=517), 56.3% reported that they experienced poultry mortality (of any quantity) within the previous 8-months (*since the Khmer New Year [April 14-16]*). The proportion of households that experienced poultry mortality differed by province ($\chi^2=25.0, p<0.001$) with the highest proportion observed in Pursat (75.9%), followed by Svay Rieng (61.8%), Banteay Meanchey (59.7%), Prey Veng (52.0%), Kampong Cham (48.2%), and the lowest in Takeo (42.9%).

4.3.4.1 Poultry morbidity and mortality

Within flock morbidity and mortality was calculated using data from the households owning poultry in Kampong Cham and Prey Veng provinces only (Table 4-4).

Table 4-4 Characteristics of reported poultry morbidity and mortality in Kampong Cham and Prey Veng, 2007

Characteristics of Mortality	Province		
	Kampong Cham (n=83)	Prey Veng (n=98)	Total (n=181) ^a
Households experiencing any poultry mortality among:			
n (%)			
Chicken Flocks	36 (43.4)	41 (41.8)	77 (42.5)
Duck Flocks	5 (6.0)	13 (13.3)	18 (9.9)
Within Flock Morbidity (=total number of birds that were ill/total flock size)*100%			
median % (IQR)			
Chicken Flocks	50.0 (24.0-83.3)	50.0 (20.0-75.0)	50.0 (24.0-75.0)
Duck Flocks	62.5 (29.1-87.5)	50.0 (25.0-77.8)	50.0 (25.0-77.8)
Within Flock Mortality (=total number of birds that died/total flock size)*100%			
median % (IQR)			
Chicken Flocks	50.0 (25.0-77.8)	50.0 (20.0-66.7)	50 (23.3-75.0)
Duck Flocks	66.7 (60.5-75.0)	50.0 (25.0-77.8)	53.6 (25.0-77.8)

^aLimited to households raising poultry only

^bIllness among chickens and ducks was evaluated according to the subject and is recognized by most villagers if their poultry suffer from seizures, have white eyes or become motionless

In Kampong Cham and Prey Veng Provinces, chicken mortality was common with 42.5% of households reporting at least one chicken having died within the previous 8-month period (since the Khmer New Year). Duck mortality was much less common with approximately 10% of households reporting that at least one duck died within the previous 8-month period. When mortality occurred in a poultry flock, approximately half of the chickens within the chicken flock (50% IQR: 24-75%) and half of the ducks within the duck flock (54% IQR: 25-77.8%) were reported to have died.

There were no linear or log-linear correlations found between mortality and flock size among chicken ($r=0.103$) or duck flocks ($r= -0.028$), or when comparing mortality $\geq 60\%$ among chicken flocks ($r=0.049$) or $\geq 30\%$ among duck flocks ($r=-0.230$).

4.3.4.1 Poultry mortality reporting

Despite 93.7% of adult respondents (2,247/2,398) believing that *it is important to report poultry mortality* and half (56%) of households experiencing any mortality, only 16.8% reported poultry mortality to the authorities (Figure 4-6). The proportion of households reporting poultry mortality differed by region with the highest reporting occurring in Svay Rieng and the lowest in Banteay Meanchey ($X^2=23.6$ $p<0.001$). No relationships were found between the proportion of households that reported poultry mortality and increasing chicken (Fishers exact $p=0.58$) or duck ($X^2=47.1$ $p=0.17$) mortality rates in Kampong Cham and Prey Veng Provinces; however these results are based on very small sample sizes (Table 4-5).



Figure 4-6 Poultry mortality vs. mortality reporting by province

Table 4-5 Within-flock mortality and poultry mortality reporting in Kampong Cham and Prey Veng provinces, 2007

Within-flock mortality*	Chicken Flocks	Duck Flocks
	n households reporting poultry mortality to authorities / n households reporting mortality ≥X%	
≥50%	7/41 (17.1)	2/11 (18.2)
≥60%	6/30 (20.0)	1/8 (12.5)
≥75%	5/21 (23.8)	0/6 (0)
100%	0/6 (0)	0/2 (0)

*Total number of birds within the household that died during the previous 8 months/ total number of birds owned during the previous 8 months

Subjects most often reported poultry mortality to village chiefs or village animal health workers (VAHW). To whom subjects reported to differed by province (Figure 4-7). For example, reporting to village animal health workers (VAHW) ranged from 9.1% in Banteay Meanchey province to 52.2% in Pursat province ($X^2=13.4$, $p=0.02$); and reporting to village chiefs ranged from 28.3% in Pursat province to 76% in Takeo province ($X^2=23.0$, $p<0.001$).

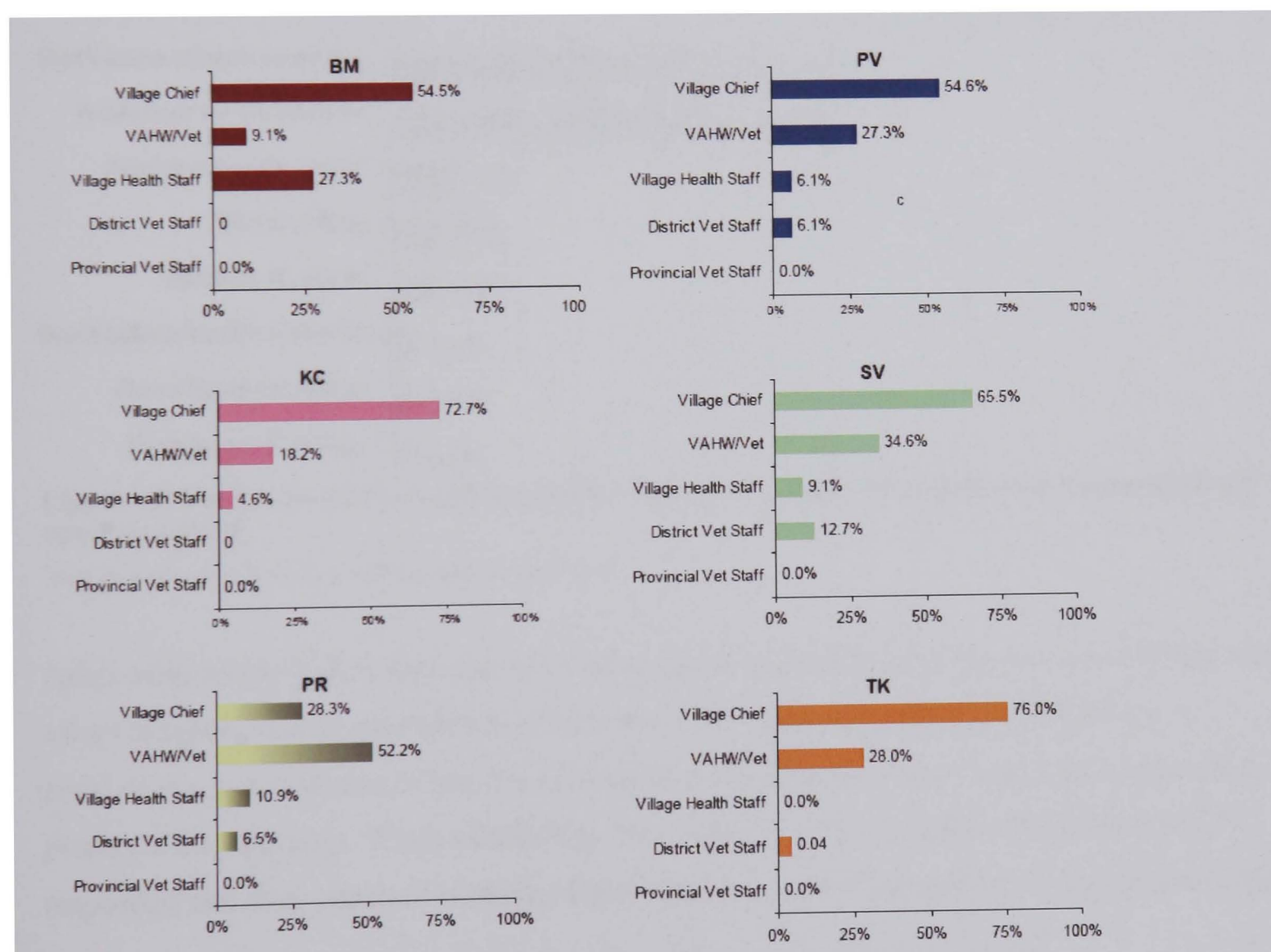


Figure 4-7 Poultry mortality reporting by province (To whom did you report?)

More than half of the adult respondents (n=2,398) felt that *AI education* (52.8%) would encourage poultry mortality reporting, while 21.7% stated that *if they were asked to report* by authorities, they would (Figure 4-8). Reasons that discouraged reporting included *not knowing where or to whom to report* (26.8%), that *poultry are not as important as compared to cattle/water buffalo* (21.6%), and that they *keep poultry to eat* (10%). Very few respondents feared culling or panic in the village as a result of reporting. Other responses to this open question are provided in Figure 4-8 below.

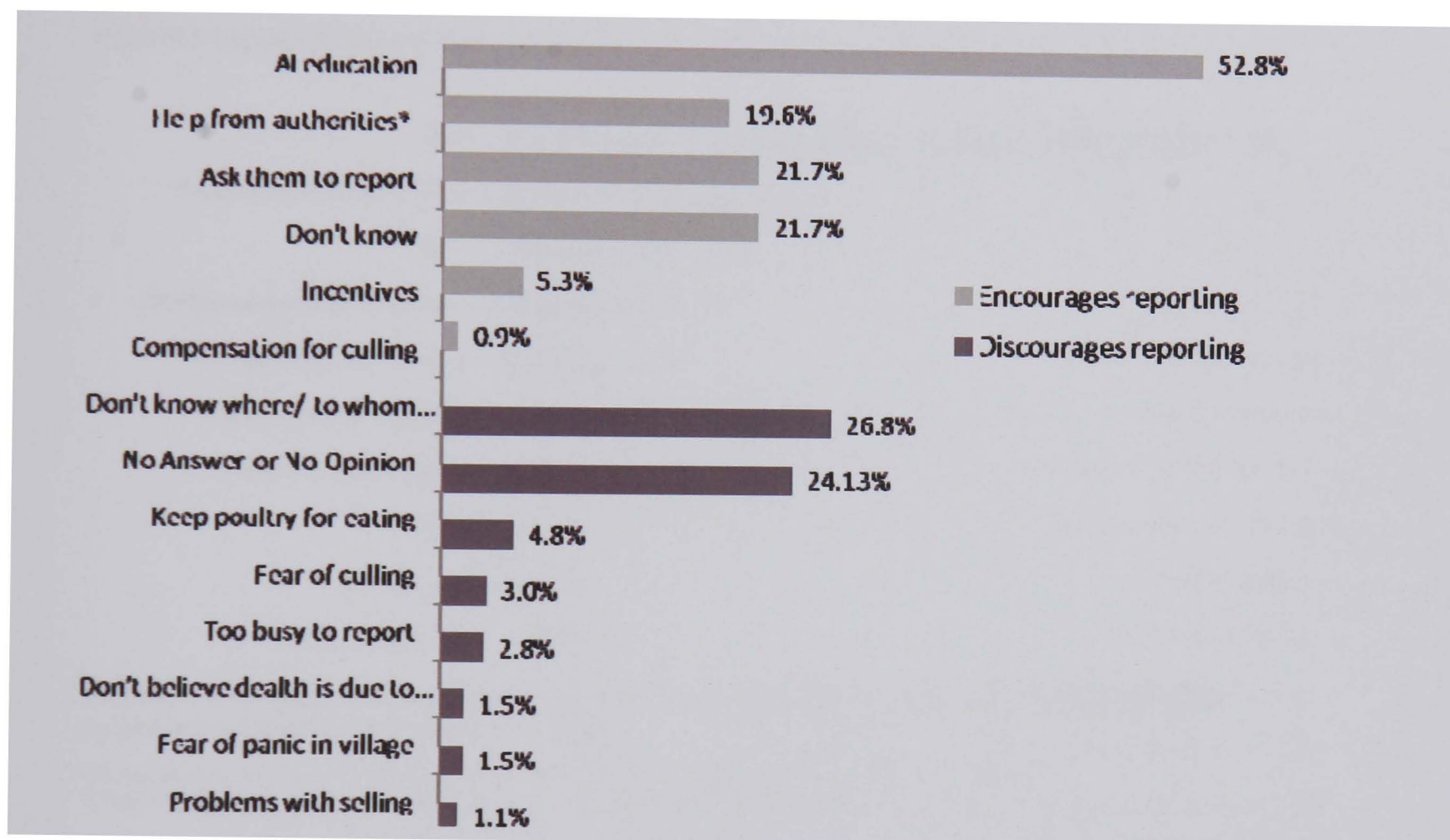


Figure 4-8 Poultry mortality reporting (What would encourage/discourage you from reporting? open question)

*Help from authorities in the form of drugs, food, and/or training

Adult subjects (n=2,400) were asked via open-ended questions what they expected if they were asked to report poultry mortality to authorities; 12% expected *medicine for poultry to be provided by veterinarians*, while 5% mentioned that authorities would *check for symptoms in poultry* after reporting. When asked what they expected if their poultry were culled, 46.2% responded that they expected *nothing*, while more than half of the subjects expected some help

from authorities either by *replacing poultry* (25.4%) or *providing money to replace poultry* (31.8%)

Eighty-eight percent of poultry owners (n=1,325⁸) reported that they buried animals that died from illness, 29.1% prepared dead poultry as food for their family, 12.1% throw dead poultry into water sources, 17.8% throw in areas around the house yard and 21.6% burned dead birds. Few subjects reported having prepared poultry that had died from illness for sale (<1%) or used them for feed for other animals (5.5%). Figure 4-9 shows the responses of subjects that had reported experiencing poultry mortality since the Khmer New year (n=1,325).

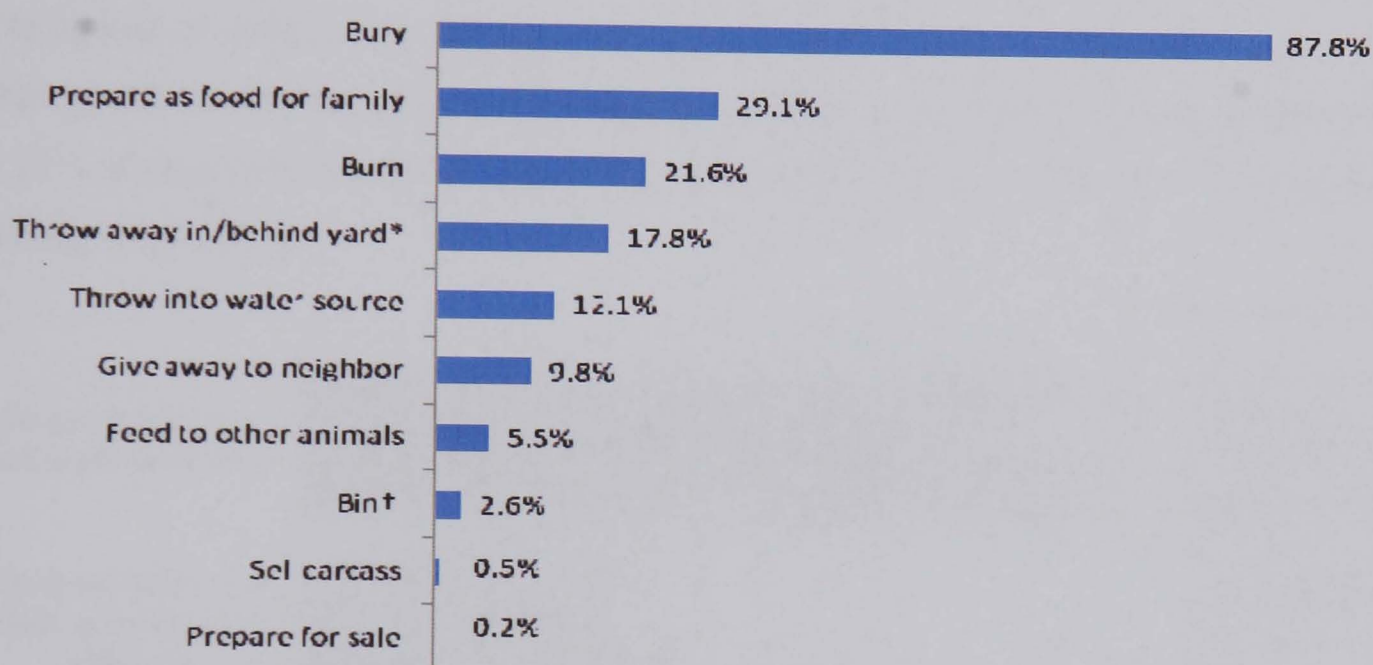


Figure 4-9 Practices with poultry that died from illness (restricted to subjects that had experienced poultry mortality, n=1,325)

*In unsafe areas, e.g., in yard, behind house yard (not in water sources); not intended to feed other animals

† Significantly higher among adult females (p<0.05); no other gender differences found

Sick poultry were treated similarly to poultry that died in that 26.8% prepared sick poultry for food for family, however 47.6% of respondents reported quarantining poultry (duration not recorded) from other household animals (e.g., pigs, dogs, other birds), 26.8% prepared poultry that died from illness for household consumption and 8.9% sold sick poultry alive.

I separately evaluated what poultry owners did with the animals in the sick flock that did not become ill (i.e., the remaining healthy flock). The majority of adult respondents reported

⁸ 1,325 indicate the total number of subjects (adults + heads of household) that owned poultry since the Khmer New Year

preparing these animals for food (73.6%), 50% sold poultry and 41.1% reported quarantining the flock from other animals (duration of quarantine was not recorded).

4.3.5 Avian Influenza and Poultry Flocks

Sixty-nine percent of adults believe that their poultry flocks are at risk of infection from AI and 30% believe they can tell if their flock is infected with H5N1 (Figure 4-10). Respondents felt that they knew AI symptoms better if AI were affecting chickens (66-86% depending on province) than ducks (22-42% depending on province), however most symptoms that they believed were due to AI are common to other poultry infections (e.g., Newcastle Disease) with the exception of sudden death and death in large numbers. Twenty-six percent thought that *sudden death* and 4% thought *death in large numbers* were symptoms of AI in chickens; 41% and 15% of respondents believed *sudden death* and *death in large numbers* were AI symptoms in ducks, respectively.

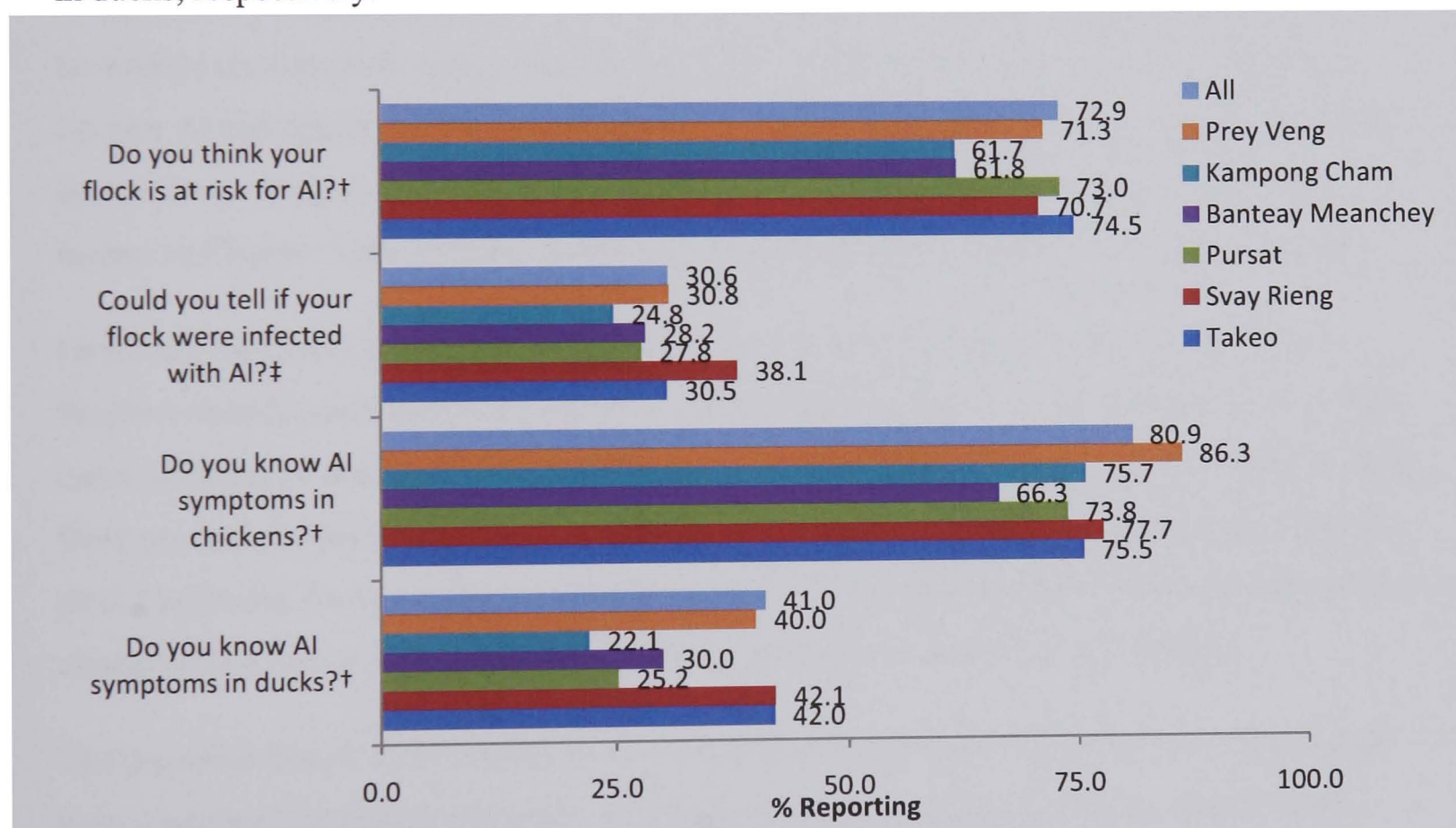


Figure 4-10 Perception of risk in domestic poultry by adults (n=2,398)

[†] X² test p<0.001; [‡] X² test p=0.002

4.3.6 Economic Importance of Poultry Rearing in the Household

Among heads of household (n=600), 29.2% (n=175) reported that their income was primarily based on poultry raising, and another 43% reported that they generated some income from raising poultry. The median annual income generated from poultry raising was reported as 100,000 riel/year (\$25/year; range= 20,000- 1,000,000 riel [\$5-250]/year; n=174).⁹ When asked how their household income would be affected if they stopped selling poultry, 32% reported it would decrease most of their income while 22% reported it would decrease their income by half.

4.4 Discussion

Most rural households in the study areas keep small flocks of less than a dozen chickens and/or ducks with outdoor access at least during part of the day. More than 90% of Cambodia's backyard poultry holdings consist of small chicken holdings (<50), which are reared for household consumption, rather than sold into the poultry market chain (discussed further in Chapter 6) and despite the fact that most flocks are allowed to roam freely to forage for food, the flocks in villages may not be dense enough to sustain transmission. This is discussed further in Chapter 6 after presentation of the results from the second cross-sectional survey.

Exploring the diversity of poultry holdings in the study population that fall within FAO's Sector 4 classification and the newly proposed subcategories within Sector 4 has allowed for a more informative description of the diversity in poultry holdings in Cambodia. Although using these proposed definitions allowed explanation of a small proportion of poultry ownership in rural Cambodia, I suspect that this would vary by country as there may be heterogeneity in the characteristics of poultry ownership in other countries with large Sector 4 holdings.

The proposed Sector 4, Sub-category A—which contains flocks of predominantly ducks and have a minimal level of biosecurity—and Sector 4, Sub- category B—which contain flocks predominantly of ducks that are allowed to forage for food throughout the village and

⁹ UNFPA estimates that the “annual average per capita income of Cambodia is less than \$290” UNFPA. 2008 Background on Cambodia. Website: <http://www.unfpa.org/focus/cambodia/background.htm>. Last accessed 24 October 2008.

sometimes in rice fields, mix with domestic animals and have the potential to mix with wild birds—have the greatest potential for sustaining H5N1 transmission in rural areas of Cambodia (Gilbert et al. 2006; Gilbert et al. 2007). A sentinel duck surveillance program that began in 2006 of this exact category of ducks had to be stopped after only one month because H5N1 was found circulating in the flocks and all duck flocks were subsequently culled (as discussed in Section 2.5.1.3) (IPC Unpublished Data).

Wild bird mixing does not seem to be a major risk factor for animal-to-animal transmission either from wild birds to domestic flocks or vice versa as the most common species of wild bird that was reported to mix with domestic flocks was the singing bird. However approximately 20% of poultry owners reported that their domestic flocks commonly mix with egrets, a species that has recently been identified as infected with H5N1 in Hong Kong (OIE 2008d). The predominance of free-ranging poultry flocks in rural areas of Cambodia requires further evaluation into the extent domestic poultry flocks forage for food outside of their home village, including more detailed information about the specific location(s) and the frequency with which they roam, the extent of village overlap of free-ranging animals, the extent free ranging birds frequent large ponds and lakes within Cambodia and the extent to which this changes during the rainy and dry seasons. This study found very little wild bird mixing among households living in the three included districts in Pursat Province along the Tonle Sap lake, reportedly due to changes in the frequency of wild birds stopping over on the lake throughout the year (MAFF Unpublished Data).

Forty three percent and 10% of households reported mortality at any level within their chicken or duck flocks, respectively. Of note is that the upper range of within-flock mortality often exceeded 50% - 75%—which would indicate that these birds might have died under suspect conditions; conditions that should have been reported to village animal health workers (VAHW) as part of Cambodia's current passive HPAI surveillance program.

Since episodes of high poultry morbidity and mortality in backyard poultry flocks are not uncommon and because it was difficult for study subjects to distinguish symptoms of AI from other poultry diseases (e.g., Newcastle disease), rural poultry owners should be instructed to report *any* and *all* mortality to individuals who have been trained to have a greater

understanding of the disease in animals (i.e., trained VAHW). Because other poultry diseases can have morbidity and mortality rates as high as 90% or 100% (e.g., Newcastle Disease (OIE 2008b)), it should be the responsibility of VAHW and NaVRI who are trained to recognize key diseases such as avian influenza and Newcastle disease in poultry to determine whether the flock died under suspect conditions (e.g., high mortality, rapid onset of death) that warrant further reporting and follow-up. Without VAHW, officials from NaVRI may be called upon to investigate numerous instances of poultry mortality that may not be necessary. Focusing on high mortality events above a certain threshold, possibly events with greater than 60% within flock mortality, would help officials differentiate common poultry mortality from more suspect mortality events. For example, using data from this study, if a threshold of 60% within flock mortality were assumed to be the basis for which VAHW called upon ministry officials to investigate, 38 households would have been visited within the 8-month study period to investigate 30 instances of >60% mortality within a chicken flock and 8 occurrences of >60% mortality within a duck flock.

This study found less than 20% of those that experienced poultry mortality reported this to the authorities and no increase in reporting occurred with increasing mortality rates. Only 17.1% (7/41) and 18.2% (2/11) of households that experienced more than 50% mortality within their chicken or duck flocks, respectively, reported this to the authorities. Only 23.8% (5/21) and 0% (0/6) of households that experienced more than 75% mortality in their chicken or duck flocks, respectively, reported this to the authorities. Although reporting of mortality was low, when it did occur villagers reported to their village chief and thus village chiefs should be included in AI education training sessions that are frequently run by FAO and NaVRI and if possible special training sessions should be held with village chiefs as well as VAHW.

Surprisingly, less than half of the study subjects stated that they did not expect any compensation from authorities when poultry died, despite approximately one third of households claiming their income is primarily based on poultry raising. Compensation from the government is not provided to individuals who have had poultry flocks culled as a result of H5N1 infection. It is possible that rural Cambodians are unaware that compensation is offered in neighbouring countries.

This study found that the use of biosecurity in animal raising should be improved to a minimum level by restricting poultry to be free ranging in specified areas (e.g., fenced in rice fields, making home fences sealant to animals) or by separating human areas from poultry. A reduction in free ranging poultry flocks, domestic/wild species mixing and domestic poultry and increase in the practice of separating domestic animals (poultry species, pigs, other animals) could not only result in the potential reduction of H5N1 poultry-to-poultry transmission but may also result in a reduction of overall poultry mortality experienced by the households.

It would be valuable to examine how the results (e.g., poultry ownership, husbandry practices, poultry mortality experienced and reported) of this study differ from other countries in the Mekong Delta Region; in particular, to compare these results with other countries with predominant Sector 4 holdings (e.g., Vietnam, Laos, Myanmar, Indonesia, Malaysia, etc) and across Africa (e.g., Nigeria (Adene & Oguntade 2006), Mali). These sub-categories may be helpful in describing the poultry production system where H5N1 domestic poultry outbreaks have been frequent (e.g., Vietnam and Thailand) to assess if there is an association between the poultry sector, and therefore husbandry practices of backyard poultry flocks, and the frequency of H5N1 outbreaks.

Backyard poultry rearing is very important to the livelihood of Cambodians as well as individuals in many countries throughout the world. However, since backyard poultry are an important element for the sustainability of HPAI infection, this type of poultry system makes controlling HPAI extremely challenging (Cecchi et al. 2008; Cristalli & Capua 2007; Songserm et al. 2006). Effective control measures would benefit from a better appreciation of the heterogeneity within this Sector, which is missed by FAO's classification. My results provide detailed information on this key sector and provide better understanding of how heterogeneous backyard poultry holdings can be. These heterogeneities should be considered when designing HPAI control strategies.

Chapter 5 Frequency and Patterns of Contact with Domestic Poultry and Potential Risk of H5N1 Transmission to Humans Living in Rural Cambodia

5.1 Introduction

Several epidemiologic studies have evaluated the risk of transmission of HPAI from poultry-to-humans (Apisarnthanarak et al. 2005; Bridges et al. 2000; Bridges et al. 2002; Hinjoy et al. 2008; Katz et al. 1999; Lu et al. 2008; Ortiz et al. 2007; Schultsz et al. 2005; Thanh Liem et al. 2005; Vong et al. 2006; Vong et al. 2009). These studies have identified several risk factors that may be associated with infection including close direct contact with poultry and indirect transmission via the environment. However, despite frequent and widespread contact with poultry, transmission from poultry to humans is rare.

Transmission of HPAI/H5N1 from poultry-to-human is most likely to occur through direct contact with aerosolized virus, infected poultry organ tissue, blood, nasopharyngeal secretions, or faeces under poor hygienic conditions, or possibly through the ingestion of contaminated water (WHO 2006g; Writing Committee of the Second World Health Organization Consultation on Clinical Aspects of Human Infection with Avian Influenza 2008). The risk of transmission will be influenced by the nature and frequency of contact with contaminated cells, tissue, blood or secretions in which the virus replicates (Beato et al. 2007; Isoda et al. 2006; Mase et al. 2006). Most of the H5N1 laboratory confirmed human cases to date have reported recent contact with infected poultry although the specific nature of the contact was not recorded (WHO 2006-2009).

Direct routes may include contact with infected blood or bodily fluids via food preparation practices (Greiner et al. 2007) (e.g., slaughtering, boiling, defeathering, cutting meat, cleaning meat, removing and/or cleaning internal organs of poultry); consuming uncooked poultry products (e.g., raw duck blood) (Apisarnthanarak et al. 2005; Beigel et al. 2005; de Jong et al. 2005) or through the care of poultry (either commercially or domestically). Little is understood

about H5N1 transmission via indirect routes, though recent studies have suggested an association between exposure to a contaminated environment (e.g., water; cleaning poultry cages or their designated areas; using poultry feces for fertilizer) and infection either through ingestion, conjunctival or intranasal inoculation of contaminated water, soil (de Jong et al. 2005; Vong et al. 2008) or via fomites on shared equipment or vehicles transporting products between farms (FAO 2004a). Other pathways may exist but are currently unknown.

At present there are an excess of reported cases in children and young adults (Ji-Ming et al. 2007). However, in the absence of detailed exposure data it is not possible to ascertain whether these represent increased exposure, susceptibility to infection, susceptibility to severe disease or a combination of all three. Case-control and seroprevalence studies have been conducted to explore risk factors for infection (Areechokchai et al. 2006; Bridges et al. 2000; Bridges et al. 2002; Dinh et al. 2006; Katz et al. 1999; Mounts et al. 1999; Ortiz et al. 2007; Schultsz et al. 2005; Thanh Liem et al. 2005; Vong et al. 2006; Vong et al. 2009). Direct contact with sick and dying poultry was noted as an important risk factor in one study (Dinh et al. 2006) but only 38% of the population risk of AI could be attributable to this exposure because of the relatively low prevalence of reporting of this practice. The power of these studies is limited because of their small sample size and there is a lack of reference data on how preparation of sick and dying poultry and other potential exposures differ within and between countries.

As described in Chapter 2, H5N1 outbreaks have been recurrent in domestic poultry and humans in Cambodia since 2004, mainly in villages located in Southern Cambodia (Buchy et al. 2007). Since household ownership of backyard poultry (FAO Sector 4 poultry production) in rural Cambodia is high (Chapter 4) an understanding of the extent and frequency of poultry handling behaviours in backyard poultry farming settings is necessary to assess the risk associated with different practices and formulate sensible recommendations to mitigate this risk. Data collected from my first cross-sectional survey was therefore used to explore patterns of human contact with poultry among rural Cambodians to identify populations with the highest H5N1 (and other HPAI) exposure potential.

5.1.1 Objectives of the Chapter

The objectives of this chapter are to:

- Describe the extent and frequency of poultry handling behaviours of rural adult males, adult females and children and how they differ by age and gender;
- Describe the understanding of avian influenza of adults and children; and
- Use risk assessment methods and the study subjects patterns of contact with poultry to generate risk indices of potential H5N1 transmission to different populations in contact with poultry.

5.2 Methods

5.2.1 Risk Assessment Framework

A conceptual pathway was developed within the risk assessment framework (1999; OIE 2005a) describing the steps to infection with H5N1 in humans from contact with poultry (Figure 5-1). The pathway includes the probability that an animal is infected with H5N1 (P), the probability that an individual comes in contact with an infected animal (C), and the probability of effective transmission of H5N1 from poultry to human in the absence of protective clothing (β).

Several important data gaps and uncertainties currently exist—namely the persistence of H5N1 in domestic/wild poultry populations and in the environment under different atmospheric conditions, virus survival in poultry meat, organs, tissue and blood, exposure quantification of H5N1 from poultry, and empirical data on risk factors for transmission from poultry to humans—making it difficult to perform a complete quantitative risk assessment (FAO et al. 2007; Greiner et al. 2007) at this time. In this analysis, my field data are used to help with such an assessment focusing on the modules outlined in bold (patterns of contact that could result in effective transmission).

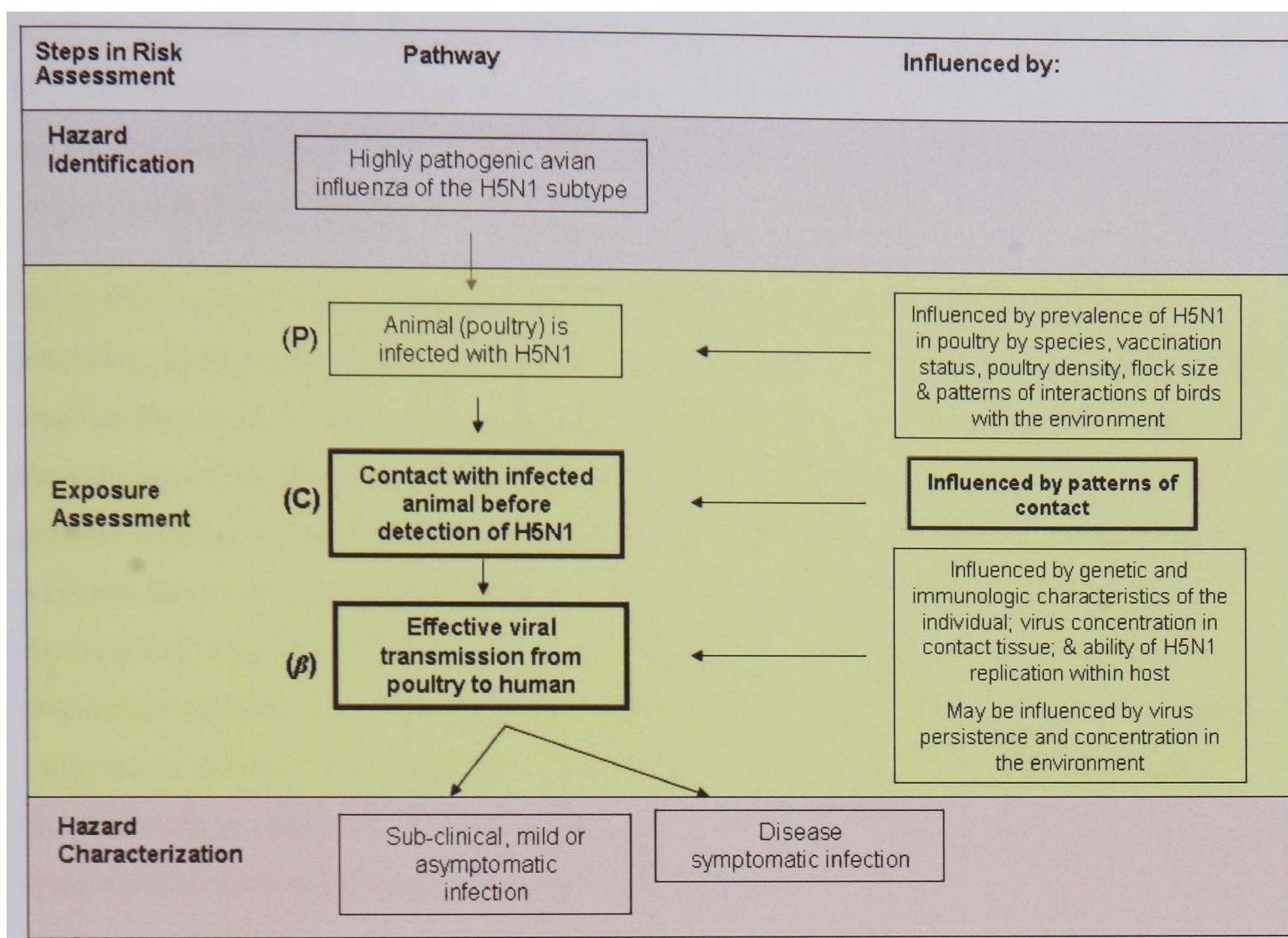


Figure 5-1 Conceptual pathway for transmission of H5N1 from poultry to humans via contact with poultry

5.2.2 Statistical Methods

5.2.2.1 Prevalence of poultry handling behaviours

Poultry contact patterns were analyzed by gender and age using non-parametric tests (chi-square tests or Fisher's exact tests) as appropriate (as mentioned Chapter 3).

5.2.2.2 Principal components analysis of food preparation variables

To reduce the number of variables that describe contact with poultry, principal component analysis (PCA) was undertaken (STATA v10). Principal component analysis is a non-parametric method of reducing the number of variables in a dataset that describe similar practices by extracting the factors that account for the most variance in the responses of the study subjects. Since there are several stages of food preparation (e.g., boil poultry, slaughter

poultry, cut poultry meat, wash poultry meat, remove internal organs and wash internal organs), PCA can determine the best possible combination of these variables (called components) that explain the overall observed variation in the responses on food preparation practices (Kirkwood & Sterne 2003).

Using PCA, a set of eigenvalues and eigenvectors are calculated for each of the factors describing food preparation (i.e., cook poultry, boil poultry, slaughter poultry, cut poultry meat, wash poultry meat, remove internal organs and wash internal organs). Eigenvalues represent the variance of the principal components where the first principal component accounts for the greatest variance in practices between individuals in the study, the second principal component accounts for the second greatest variance, and so on. For each principal component, a set of eigenvectors or coefficients ($\alpha_{n1}, \alpha_{n2}, \alpha_{n3}, \alpha_{n4}, \alpha_{n5}, \alpha_{n6}, \alpha_{n7}$) exist for each of the 7 food preparation variables. Scree plots (Cattell 1966) are used to determine the number of principal components to retain. New variables representing each component (principal component 1 (PC₁), principal component 2 (PC₂) ... principal component n (PC_n)) are created for each subject in the study using eigenvectors as weights, where:

$$\begin{aligned}
 PC_1 &= \alpha_{11}X_{11} + \alpha_{12}X_{12} + \alpha_{13}X_{13} + \alpha_{14}X_{14} + \alpha_{15}X_{15} + \alpha_{16}X_{16} + \alpha_{17}X_{17} \\
 PC_2 &= \alpha_{21}X_{21} + \alpha_{22}X_{22} + \alpha_{23}X_{23} + \alpha_{24}X_{24} + \alpha_{25}X_{25} + \alpha_{26}X_{26} + \alpha_{27}X_{27} \\
 &\dots \\
 PC_n &= \alpha_{n1}X_{n1} + \alpha_{n2}X_{n2} + \alpha_{n3}X_{n3} + \alpha_{n4}X_{n4} + \alpha_{n5}X_{n5} + \alpha_{n6}X_{n6} + \alpha_{n7}X_{n7}
 \end{aligned}$$

Where X_{n1} =cook poultry, X_{n2} =boil poultry, X_{n3} =slaughter poultry, X_{n4} = cut poultry meat, X_{n5} =wash poultry meat, X_{n6} =remove internal organs and X_{n7} =wash internal organs. These newly created variables (PC₁ and PC₂) were subsequently analyzed by gender and age group using t-tests or Wilcoxon rank-sum tests as appropriate.

5.2.2.3 Estimates of human exposure risk

Risk profiles were generated for each subject using their individual poultry handling contact patterns. The probability of effective viral transmission following a certain type of contact is assumed to be high, moderate or low as indicated in Table 5-1. A transmission risk weighting score (β) was applied to quantify the risk associated with high and moderate risk practices

compared to low risk practices. Practices listed in group 1 are believed to have a higher potential transmission risk based on the nature of contact and potential H5N1 exposure than practices listed in 2 or 3 while practices listed in group 2 have a higher potential transmission risk than practices in group 3. In the analysis presented here, I used values $\beta_1 = 10$, $\beta_2 = 2$ and $\beta_3 = 1$. The values I chose for β_1 , β_2 and β_3 are used in this analysis as an illustration of weighting exposures and are based on available data on the pathogenicity of H5N1 in poultry tissues (Beato et al. 2007; Das A 2008; FAO 1999; Hulse-Post et al. 2005; Mase et al. 2006; Pantin-Jackwood et al. 2007; Perkins & Swayne 2001; Swayne & Beck 2005; Swayne 2007; Webster et al. 2007; WHO 2006g). As more epidemiologic and virologic data about the persistence of H5N1 in poultry are collected, more precise estimates for the β values may become available.

Estimates of human exposure risk for each study participant (n=3,600) were then obtained by multiplying each reported practice with the transmission risk-weighting factor and summing these over all practices reported by each individual ($\sum \beta C$). In addition, sensitivity analyses were conducted by varying the weightings of β_1 , β_2 and β_3 (e.g., $\beta_1 = 1$, $\beta_2 = 1$ and $\beta_3 = 1$; and $\beta_1 = 20$, $\beta_2 = 5$ and $\beta_3 = 1$) to determine the effect on the human exposure risk scores.

The exposure risks were analyzed by age and gender using t-tests or Wilcoxon rank-sum tests as appropriate. P-values of <0.05 were considered statistically significant. All statistical analyses were performed using STATA (v.10) (StataCorp, College Station, Texas).

5.3 Results

5.3.1 Poultry Handling Behaviours of Adults and Children

Contact patterns with domestic poultry are provided in Table 5-1.

Table 5-1 Prevalence of practice associated with poultry in rural Cambodian households, main sources of potential exposure and weighted transmission risk potential (β)

Probability of effective viral transmission (β Grouping)	Practice	n (%)			p-value		Potential Viral Exposure
		Adult Males (n=1,201)	Adult Females (n=1,199)	Children (n=1,200)	Adult males vs. Adult females	Adults vs. Children	
		>15 yrs old		≤15 yrs old			
High (β1)	Remove internal organs (poultry)	733 (61.0)	588 (49.0)	156 (13.0)	<0.001	<0.001	O, B
	Blow into beak (FC)	19 (1.6)	1 (0.1)	6 (0.5)	<0.001	0.27	NS, B
	Kiss, suck, lick wounds (FC)	10 (0.8)	0 (0)	6 (0.5)	0.002	0.72	B
	Share water from the same bottle (FC)	21 (1.8)	4 (0.3)	21 (1.75)	0.001	0.07	NS, B
	Clean trachea (FC)	44 (3.7)	1 (0.1)	16 (1.3)	<0.001	0.235	NS, B
	Clean feathers (FC)	52 (4.3)	6 (0.5)	34 (2.8)	<0.001	0.46	B, F
	Wash internal organs (poultry)	745 (62.0)	775 (64.6)	249 (20.0)	0.185	<0.001	O, B
	Slaughter poultry	655 (54.5)	224(18.7)	138 (11.5)	<0.001	<0.001	B, F
Moderate (β2)	Touch/play with sick poultry or poultry that died from illness	597 (49.7)	485 (40.5)	90 (7.5)	<0.001	<0.001	B, F
	Use poultry faeces as manure	664 (55.3)	678 (56.6)	--	0.534	--	F
	Cut poultry meat	716 (59.6)	917 (76.5)	152 (12.7)	<0.001	<0.001	B
	Wash poultry meat	772 (64.3)	906 (75.6)	234 (19.5)	<0.001	<0.001	B
	Swim/bathe in water source where poultry have access [†]	56 (14.0)	41 (10.3)	196 (16.3)	0.113	0.01	F, NS
	Remove feathers from sick poultry [†]	76 (19.0)	101 (25.3)	102 (8.5)	0.04	0.001	NS, B, F
	Cleaning/sweeping poultry areas	843 (70.2)	903 (75.1)	442 (36.8)	0.005	<0.001	F
	Shopping at wet/live market for poultry	141 (11.7)	126 (10.5)	--	0.341	--	B, F
	Boil poultry	673 (56.0)	898 (74.9)	228 (19.0)	<0.001	<0.001	B, F
Low (β3)	Living in a household with poultry (raised chickens or ducks within previous 8 months)	517 (86.7) [‡]		1,039 (86.6)	--	0.81	F

Key: FC= Fighting Cocks; B=Blood, F= Faeces, NS= Nasopharyngeal secretions; O=Organ tissue; --not assessed

[†] This practice was only evaluated in adults from 2 provinces n=400 adult males and 400 adult females

[‡]Evaluated from head of household questionnaire only (n=600)

5.3.1.1 Food preparation practices

Preparing poultry for consumption consists of a series of steps including slaughtering the animal by breaking the neck or cutting the throat, bleeding, boiling, defeathering, removing and washing internal organs, and cutting and washing meat. Although family members as young as two years old reported being involved in the preparation of poultry for consumption during the study periods, these practices were primarily the responsibility of family members 16–60 years old (Figure 5-2).

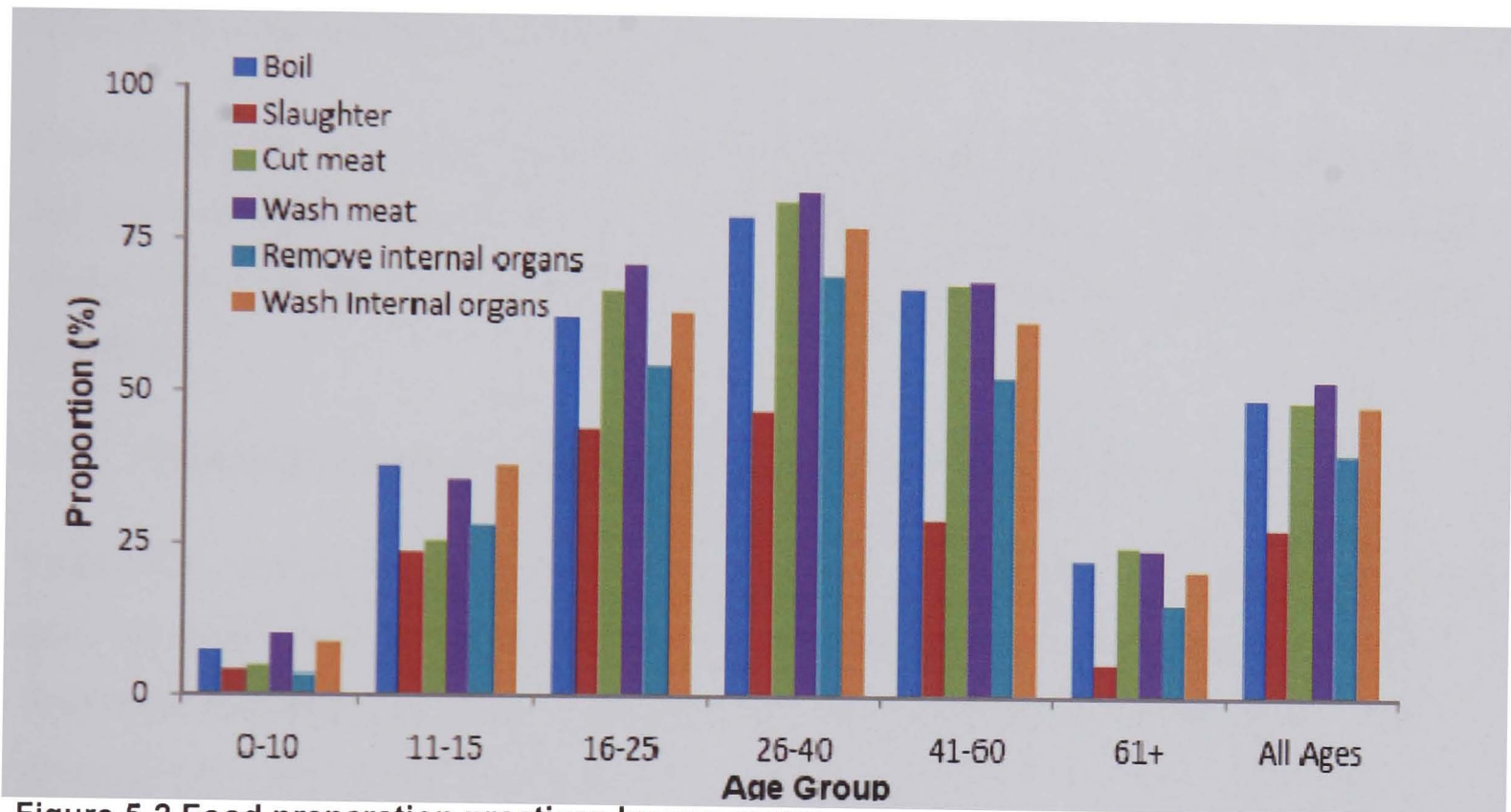


Figure 5-2 Food preparation practices by age group

Both men and women were involved in each of the stages of food preparation (Table 5-2), however overall, the proportion of adult subjects involved in all practices related to food preparation was higher than child subjects (Table 5-2). Among adults (n=2,400) significantly more men than women slaughter poultry and remove internal organs whereas adult women more often boil poultry, cut meat and wash meat.

Table 5-2 Food preparation practices by age group (n=3,600)

Practice	Adults ≥16 years old (% reporting)			χ ² (adult male v. adult female)	p-value	Children <16 years old (% reporting)		
	Total (n=2400)	Male (n=1201)	Female (n=1199)			Total (n=1200)	χ ² (adult v. child)	p-value
Slaughter/ bleed	36.6	54.5	18.7	332.3	<0.001	11.5	249.1	<0.001
Boil	65.5	56	74.9	94.4	<0.001	19	690.7	<0.001
Remove internal organs	55	61	49	34.9	<0.001	13	584.4	<0.001
Wash internal organs	63.3	62	64.6	1.7	0.185	20	601.2	<0.001
Cut meat	68	59.6	76.5	78.5	<0.001	12.7	981.3	<0.001
Wash meat	69.9	64.3	75.6	36.3	<0.001	19.5	816.6	<0.001

Among children, more males than females slaughtered poultry (17.0% vs. 5.8%, p<0.001) and removed internal organs (15.7% vs.10.2%, p=0.005), while more females than males are responsible for boiling poultry (22.3% vs.15.9%, p=0.005) and cutting meat (15.7% vs.9.8%, p=0.002).

5.3.2 Principal Component Analysis of Food Preparation Variables

Using PCA, a set of eigenvectors and eigenvalues were calculated for each of the factors (i.e., cook, slaughter, boil, remove internal organs, wash internal organs, cut meat, wash meat) describing food preparation. Two principal components were retained on the basis of the scree plot (i.e., the number of components to use in further analyses is indicated by the inflection point in the graph; Figure 5-3).

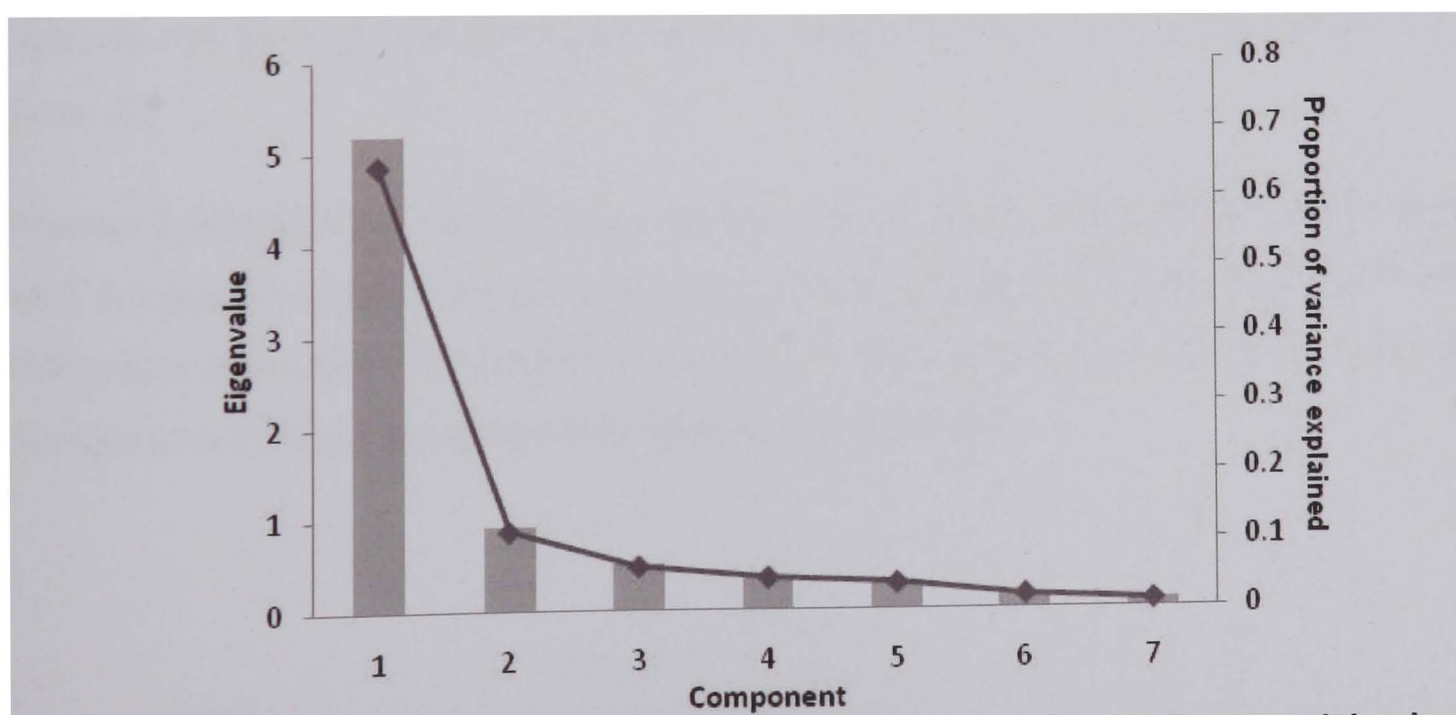


Figure 5-3 Scree plot of eigenvalue (primary axis), proportion of the variance explained (secondary axis) by principal component

The first principal component (PC₁ or practice 1), which accounts for approximately 70% of the total variation in practices between individuals in the survey, consisted of all seven of the original food preparation variables (cook, boil, slaughter, cut meat, wash meat, remove internal organs, wash organs) and hence can be interpreted as general food preparation.

The second principal component (PC₂, practice 2), which accounts for a further 12% of the variation, was dominated by the practice of slaughtering (highlighted in red in Table 5-3).

Table 5-3 Eigenvectors for each principal component

Variable	Principal Component			
	α	1	α	2
Cook	α_{11}	0.3368	α_{21}	-0.3832
Boil	α_{12}	0.3780	α_{22}	-0.3141
Slaughter	α_{13}	0.2792	α_{23}	0.7462
Cut Meat	α_{14}	0.4105	α_{24}	-0.2213
Wash Meat	α_{15}	0.4200	α_{25}	-0.1359
Remove Internal Organs	α_{16}	0.3922	α_{26}	0.3332
Wash Internal Organs	α_{17}	0.4087	α_{27}	0.1388

Two new variables were created using eigenvectors as weights (Table 5-3), where practice 1 ($PC_1 = \alpha_{11}X_{11} + \alpha_{12}X_{12} + \alpha_{13}X_{13} + \alpha_{14}X_{14} + \alpha_{15}X_{15} + \alpha_{16}X_{16} + \alpha_{17}X_{17}$) and practice 2 ($PC_2 = \alpha_{21}X_{21} + \alpha_{22}X_{22} + \alpha_{23}X_{23} + \alpha_{24}X_{24} + \alpha_{25}X_{25} + \alpha_{26}X_{26} + \alpha_{27}X_{27}$). The frequency of practice 1 (general food preparation, 71% variation) follows a similar age pattern in males and females with the highest scores between the ages of 16-25, 26-40, and 41-60 (Figure 5-4). Subjects >60 years old had lower practice scores than children between the ages of 11-15 years old.

Practice 2 (slaughtering and removing internal organs) shows greater differences by gender with this practice predominately undertaken by males (Figure 5-5). There are significant differences in practice 2 by gender among subjects with males reporting higher scores than females across all age groupings (two-sample t-test $p < 0.001$).

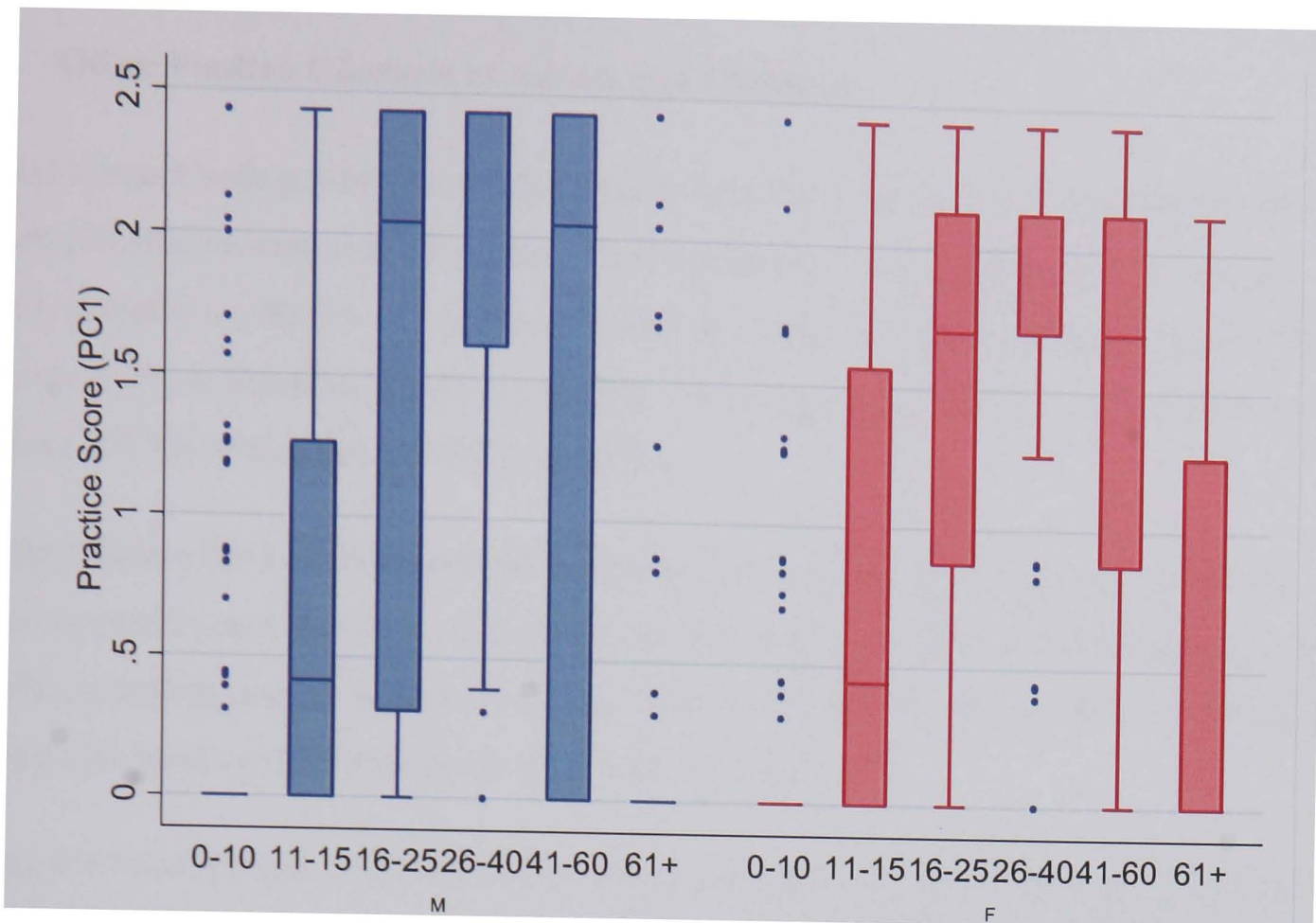


Figure 5-4 Practice 1-general food preparation-by age and gender

Note: the median value is indicated by the horizontal bar inside the box, the upper and lower edges of the box are the 75th and 25th percentiles, respectively; the * are outliers (>1.5 times the IQR); the upper and lower edges of the whiskers (lines) are the largest and smallest non-outlier values

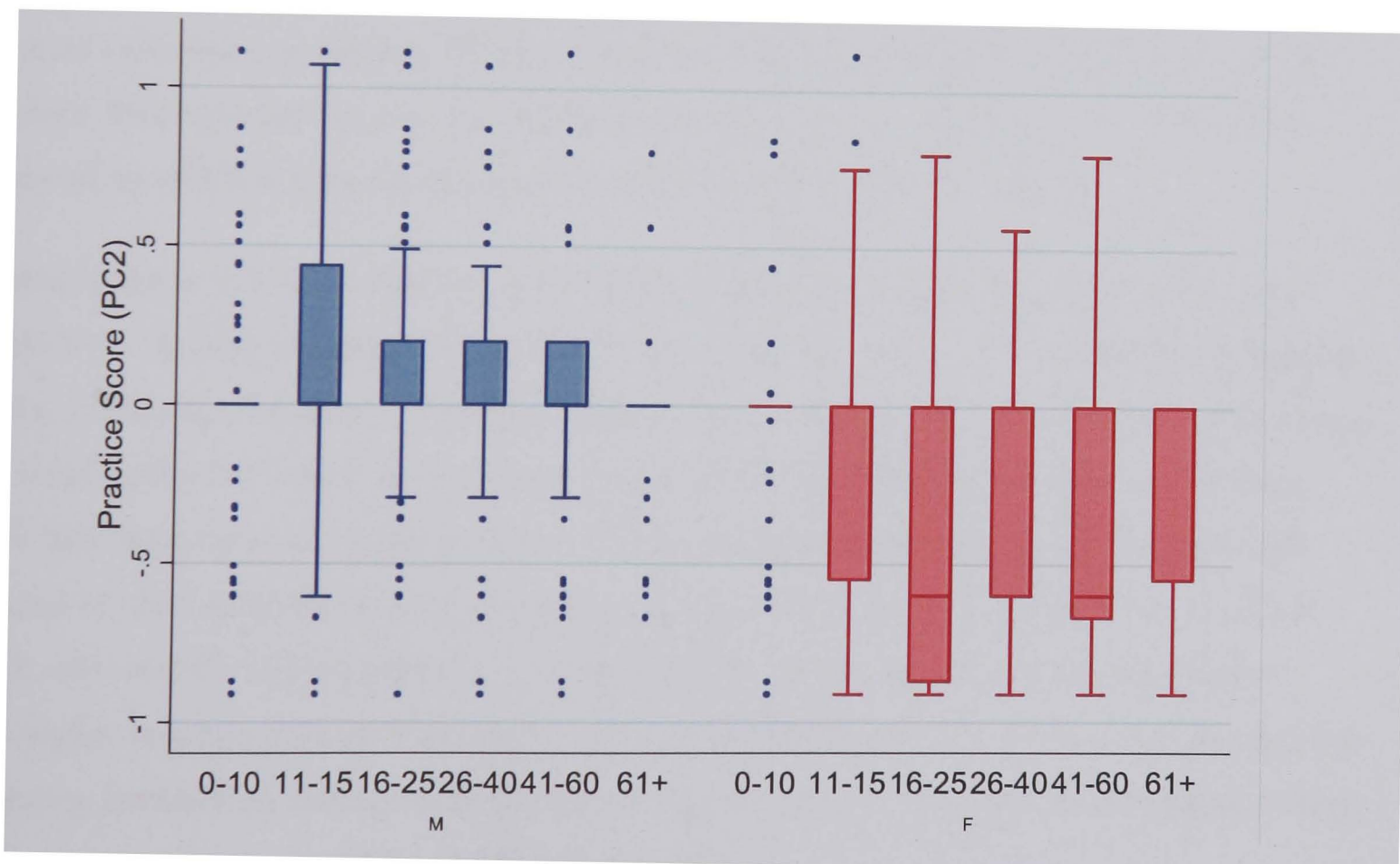


Figure 5-5 Practice 2-slaughtering and removing internal organs-by age and gender

Note: the median value is indicated by the horizontal bar inside the box, the upper and lower edges of the box are the 75th and 25th percentiles, respectively; the * are outliers (>1.5 times the IQR); the upper and lower edges of the whiskers (lines) are the largest and smallest non-outlier values

5.3.3 Other Poultry Contacts of Adults and Children

Regular contact with poultry for adult subjects (n=2,400) also includes using faeces for manure (55.9%; no variation by gender), touching sick or dead poultry with bare hands (49.7% in males vs. 40.6% in females, $X^2=20.8$, $p<0.001$), caring for fighting cocks (5.0% in males vs. 1.4% in females, $X^2=24.7$, $p<0.001$), and preparing wild birds for food (36.4% in males vs. 19.3% in females, $X^2=87.6$, $p<0.001$).

Among children (n=1,200) household responsibilities include feeding poultry (77.3%), gathering poultry and placing in designated areas or cages (43.5%), gathering/touching eggs (45.6%), cleaning poultry faeces (44.2% in males vs. 37.4% in females, $X^2=5.1$, $p=0.02$) and treating sick poultry with traditional medicines (18.5%).

Within the recall period, 35.9% of children reported that they had usually played with birds that were alive (42.5% male vs. 29.0% female, $X^2=23.8$, $p<0.001$), 2.7% reported playing with sick birds and 4.2% reported playing with dead birds (no gender difference). Thirty-two percent of children reported removing feathers from sick/dead birds (no gender difference), and 16.3% of children bathed or swam in ponds (no gender difference) in which poultry have access: of those 37.8% reported doing this every day. Twelve percent of adults (n=799) reported swimming, bathing or fishing in ponds where poultry have access (did not vary by gender). This reported activity was highest in children between the ages of 11-15 (16.5%) followed by children between the ages of 1-10 (16.2%) compared to adults.

A small number of child subjects were involved in the care of fighting cocks (5.7%; n=68; Table 5-1). Among children (n=1,200) 6.7% feed fighting cocks; 2.6% touch bloody fighting cocks; 2.8% clean feathers; 1.3% clean trachea with a swab or feather; 1.8% share water from the same bottle; 0.5% kiss, suck or lick wounds; and 0.5% blow into the beak of a fighting cock (the latter three are practices that occur during fighting cock matches). Twenty-eight percent of child subjects reported attending fighting cock matches compared with 11.3% of adult subjects ($X^2=157.9$, $p<0.001$). Among children, attendance at fighting cock matches was higher among males than females (35.0% vs. 20.6%; $X^2=30.7$, $p<0.001$). Adults reported attending matches on average once per week with the highest proportion of attendance among males between the ages of 16 and 25 years old (31.7%).

5.3.4 Use of Personal Protective Equipment when Handling Poultry

The use of personal protective equipment (e.g., gloves, rubber boots, face masks, aprons) among those that came in contact with poultry in a domestic setting was low. Few individuals had these items in their homes (as was observed by interviewers) however, more than half of the subjects reported (but were not observed) wearing such items when handling poultry (Table 5-4).

Table 5-4 Reported use of PPE by subjects caring for poultry (n=1746)

Protective Equipment	% Yes	% reporting used	
		Everyday	Sometimes
Gloves in household	7.2		
Use Gloves when touching poultry (n=125)		65.1	25.0
Boots in household	6.8		
Use boots when touching poultry (n=118)		48.3	26.7
Apron in household	7.3		
Use apron when touching poultry (n=127)		64.3	32.5
Mask in household	26.4		
Use mask when touching poultry (n=461)		61.5	25.2

[†] Restricted to adults that cared for poultry

5.3.5 Awareness of Avian Influenza

5.3.5.1 Adults

Awareness of “bird flu” is high among adults (98.9%). The primary source of information for adults (n=2,400) is TV (89%), radio (76%) and posters (20%), however significantly more adult males than adult females report hearing/learning about AI from TV (90.8% vs. 87.2%; $X^2=7.9$, $p=0.005$), radio (82.5% vs. 69.2%; $X^2=57.3$, $p<0.001$), newspapers (3.5% vs. 2.0%, $X^2=4.9$, $p=0.03$), posters (22.9% vs. 16.9%, $X^2=15.2$, $p<0.001$), village vet staff (6.4% vs. 4.1%, $X^2=6.4$, $p=0.01$), and NGO health education sessions (11.6% vs. 8.4%, $X^2=6.9$, $p=0.009$).

Figure 5-6 illustrates the differences in the source of bird flu information in the six provinces. Of special note is the large variation in the proportion of subjects that learn about AI from television (76-96% depending on province) and radio (54-84% depending on province).

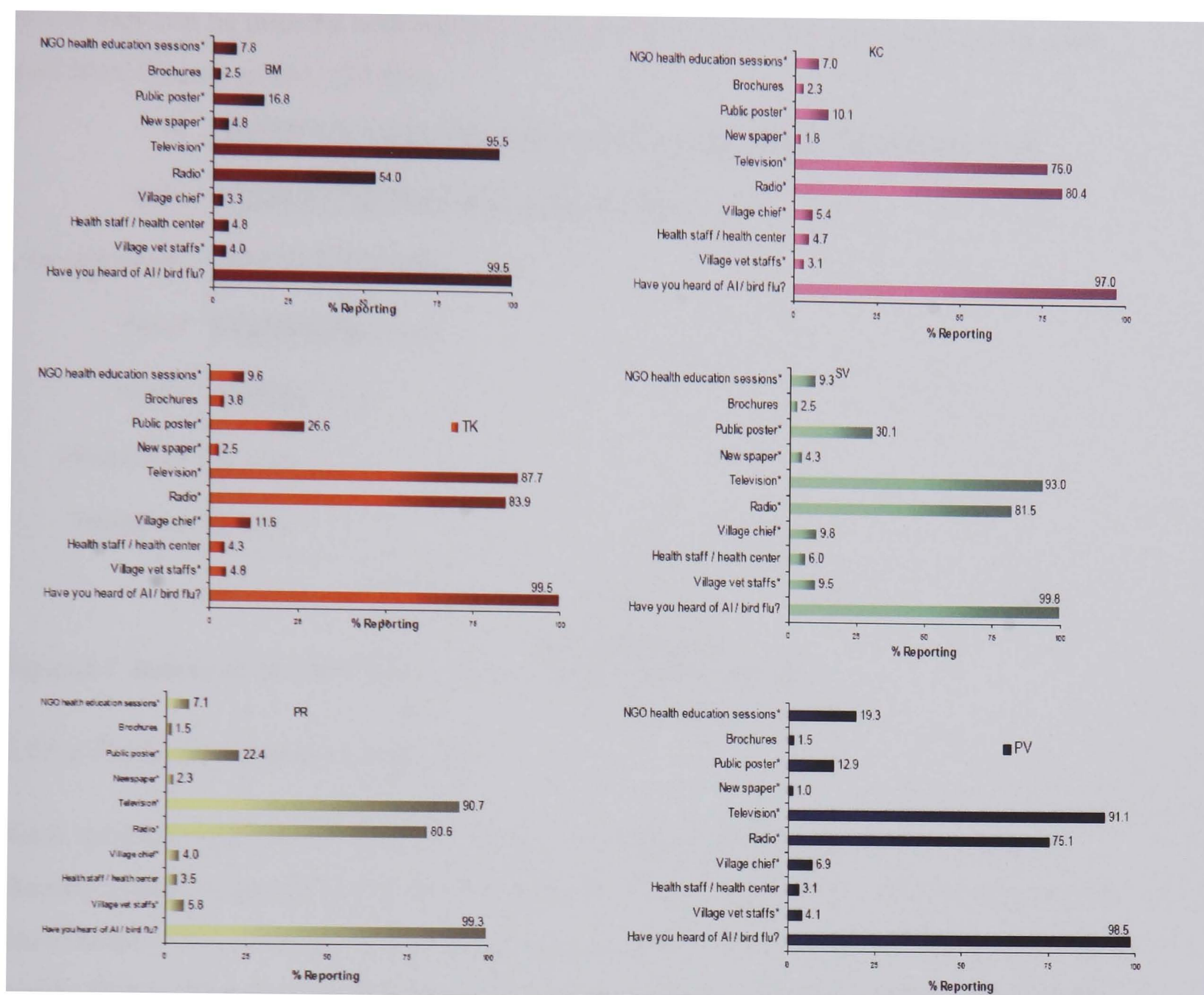


Figure 5-6 Source of AI information among adults by province (≥ 16 years old, n=2,400)

BM=Banteay Meanchey; KC = Kampong Cham; PR=Pursat; PV= Prey Veng; SV=Svay Rieng; TK=Takeo

5.3.5.2 Children

Among child respondents (n=890¹⁰), awareness of “bird flu” is high (94%); however the proportion of children that had heard of “bird flu” increased by age (68.2% 1-4 yrs old. 89.9% 5-9 years old, 98.0% 10-15 years old); $X^2=73.7$, $p<0.001$). The main sources of their AI information include TV (91%), radio (55%) and school (29%; Figure 5-7). Despite high awareness of AI, approximately half (45.8%) of the children believe that they cannot become infected with AI, however this varied by age group (29.0% 1-4 yrs old. 30.2% 5-9 years old, 52.6% 10-15 years old); $X^2=47.0$, $p<0.001$). Less than half of the children interviewed (41%)

¹⁰ Only children who were able to respond to the questions themselves (i.e., without the assistance of a parent or guardian) were asked knowledge and attitude questions about AI (n=890/1,200; 74%)

believe they can be infected with AI by touching (any) poultry or by touching sick or dead (died from illness) poultry (38.4%).

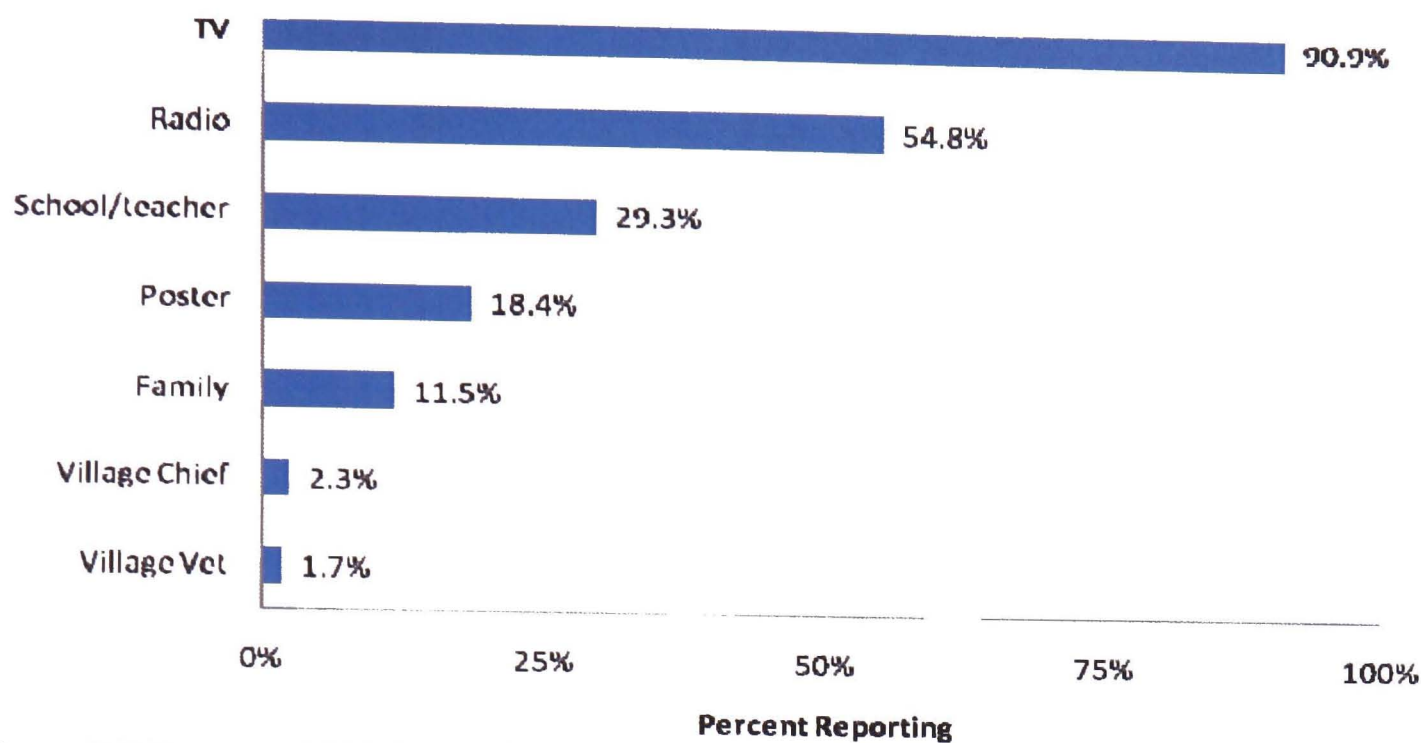


Figure 5-7 Source of AI information among children (1-15 years old)

5.3.5.3 Transmission of avian influenza

Adult subjects were asked a series of questions about possible modes of AI transmission (human-to-human, animal-to-human transmission). Their responses are shown in Figure 5-8. For example, 76% of adults believe that they can become infected with AI from swimming in ponds. Eighty-three percent of adults believe they can become infected with AI by touching poultry faeces, however this was believed by significantly more men (86%) than women (79%; $X^2=18.8$, $p<0.001$). Of note, approximately 60% of the adults surveyed believe that AI can be transmitted via sexual contact similar to HIV transmission and 89% believe that AI comes from poultry raised outside of Cambodia whereas 31% believe that AI can be found in locally raised species.

Approximately 97% of adults believe that AI can be transmitted through undercooked poultry products, while only 10% believe that AI can be transmitted via well-cooked poultry products. Seventy-four percent of adults believe risky practices include touching wild birds (74.2%), eating wild birds (76%), touching poultry blood (91%), eating eggs from healthy poultry (62.5%), touching healthy poultry (8%), touching sick or dead poultry with bare hands (96.5%), or from other people (8.2%).

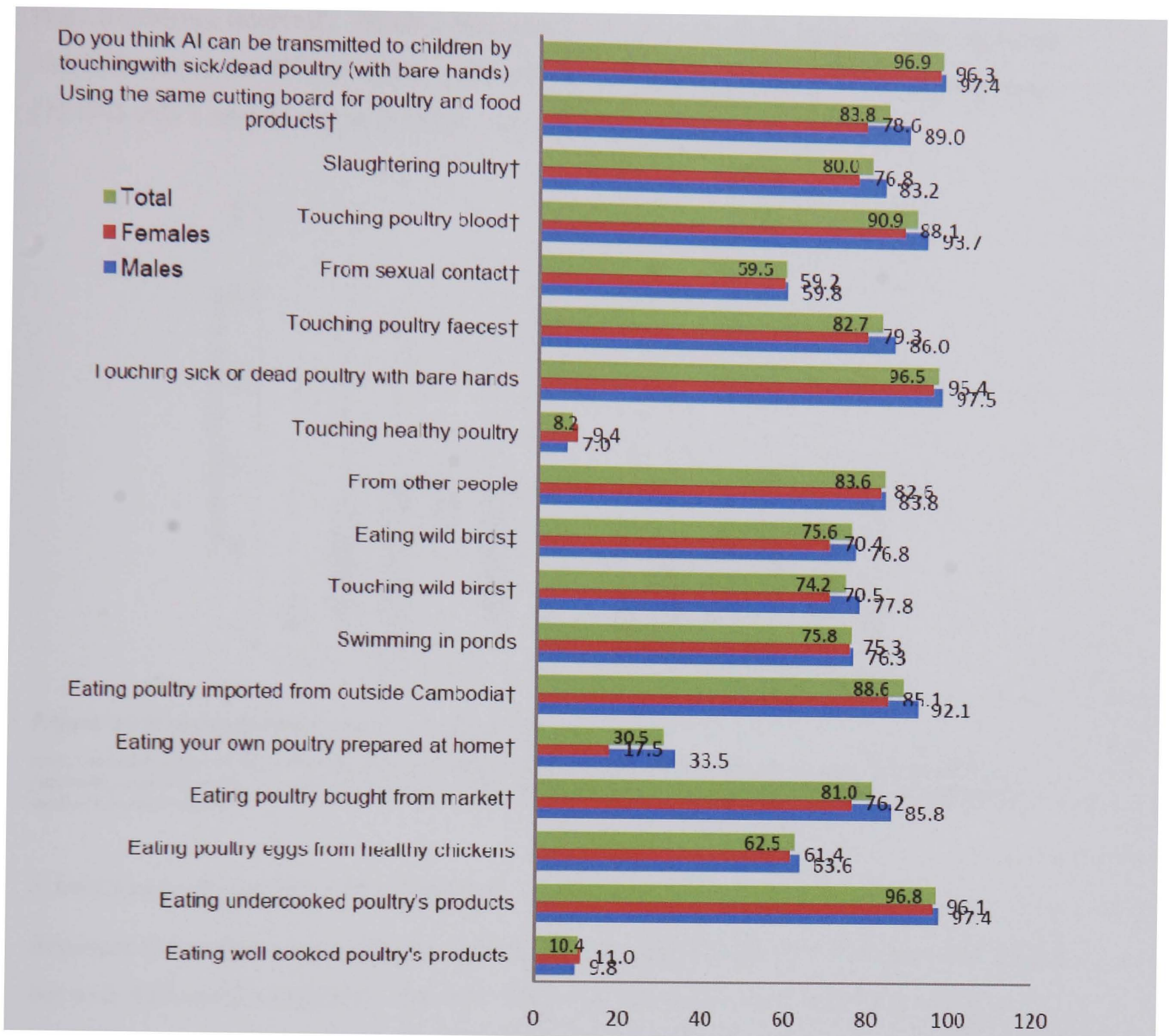


Figure 5-8 Beliefs on transmission of AI among adults (Do you think AI can be transmitted by...; n=2,398)

† X² test p<0.001; ‡ X² test p=0.001

5.3.6 Estimates of Exposure Risk

Based on the identified patterns of contact and assumptions of transmission risk (β ; Table 5-1), estimates of exposure risk were calculated for each subject and analyzed stratified by age and gender (Figure 5-9). Overall, exposure risk was higher among males than females for subjects above the age of 10 (11-15 age group, p=0.002; 16-25 age group p<0.001; 26-40 age group, p<0.001; 41-60 age group, p<0.001; 61+, p<0.001). In both males and females exposure risk varies by age with the greatest risks among males between the ages of 26-40 and 16-25 (Figure 5-9). There was also a high degree of variability in risk (as seen in the

large confidence intervals). Of the 3,600 subjects, there were 590 subjects with an exposure risk score above the 90th percentile of the sample. These subjects were predominately male (72.6%) with a median age of 30 (IQR range 21-42).

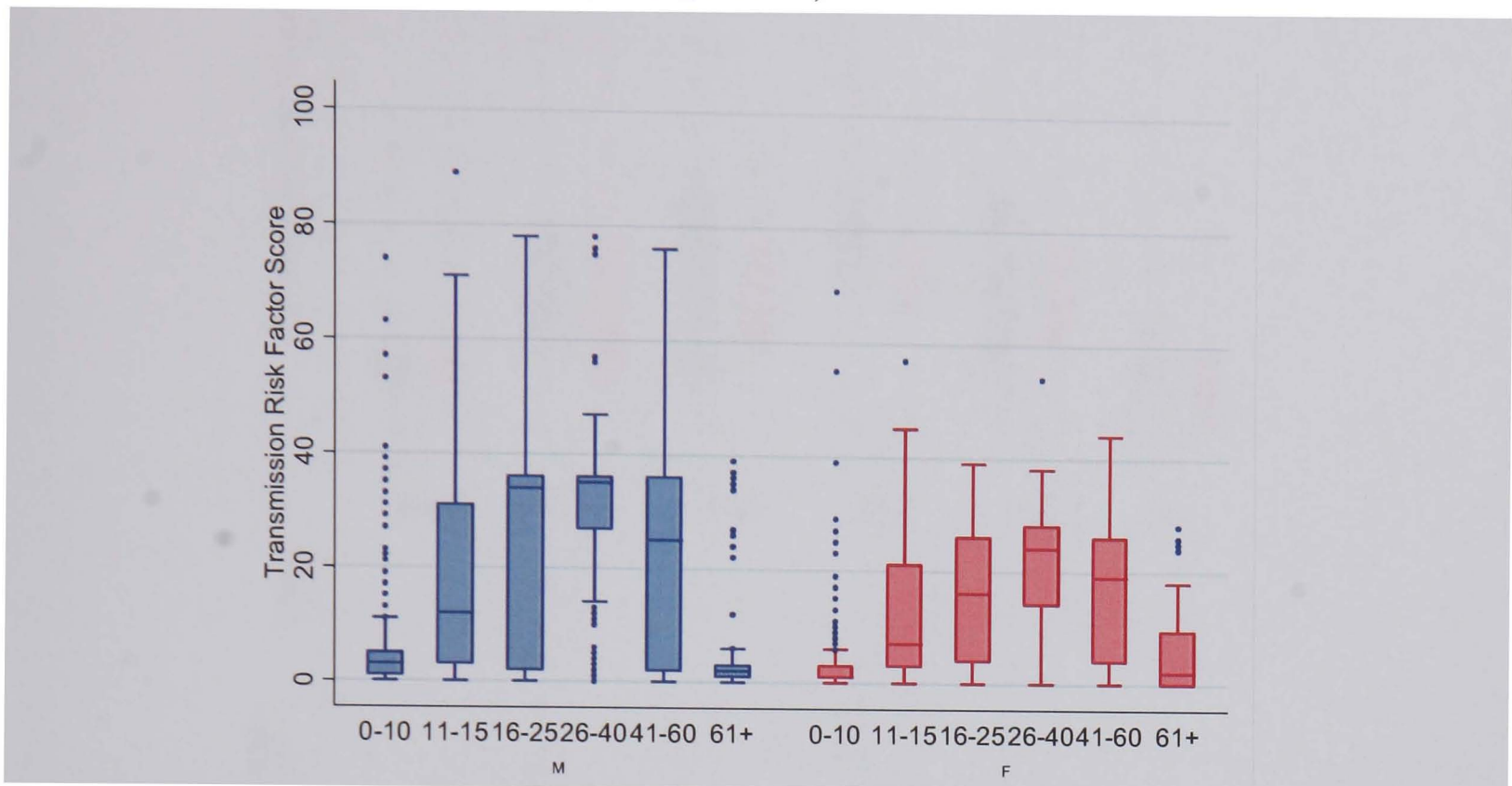


Figure 5-9 Exposure risk scores by age and gender

Note: the median value is indicated by the horizontal bar inside the box, the upper and lower edges of the box are the 75th and 25th percentiles, respectively; the * are outliers (>1.5 times the IQR); the upper and lower edges of the whiskers (lines) are the largest and smallest non-outlier values

5.3.6.1 Sensitivity analysis of β weightings

Exposure risk scores were also created using identical methods as described in section 5.2.2.2 but with different β weightings. Figure 5-10a shows the results of the exposure risk scored by age and gender using a 1:1:1 ratio for β_1 , β_2 and β_3 and Figure 5-10b shows the results of the exposure risk scored by age and gender using a 20:5:1 ratio for β_1 , β_2 and β_3 . These results find a similar risk pattern with respect to age and gender among subjects. The highest risk scores are among males between the ages of 26-40 followed closely by males between the ages of 16-25.

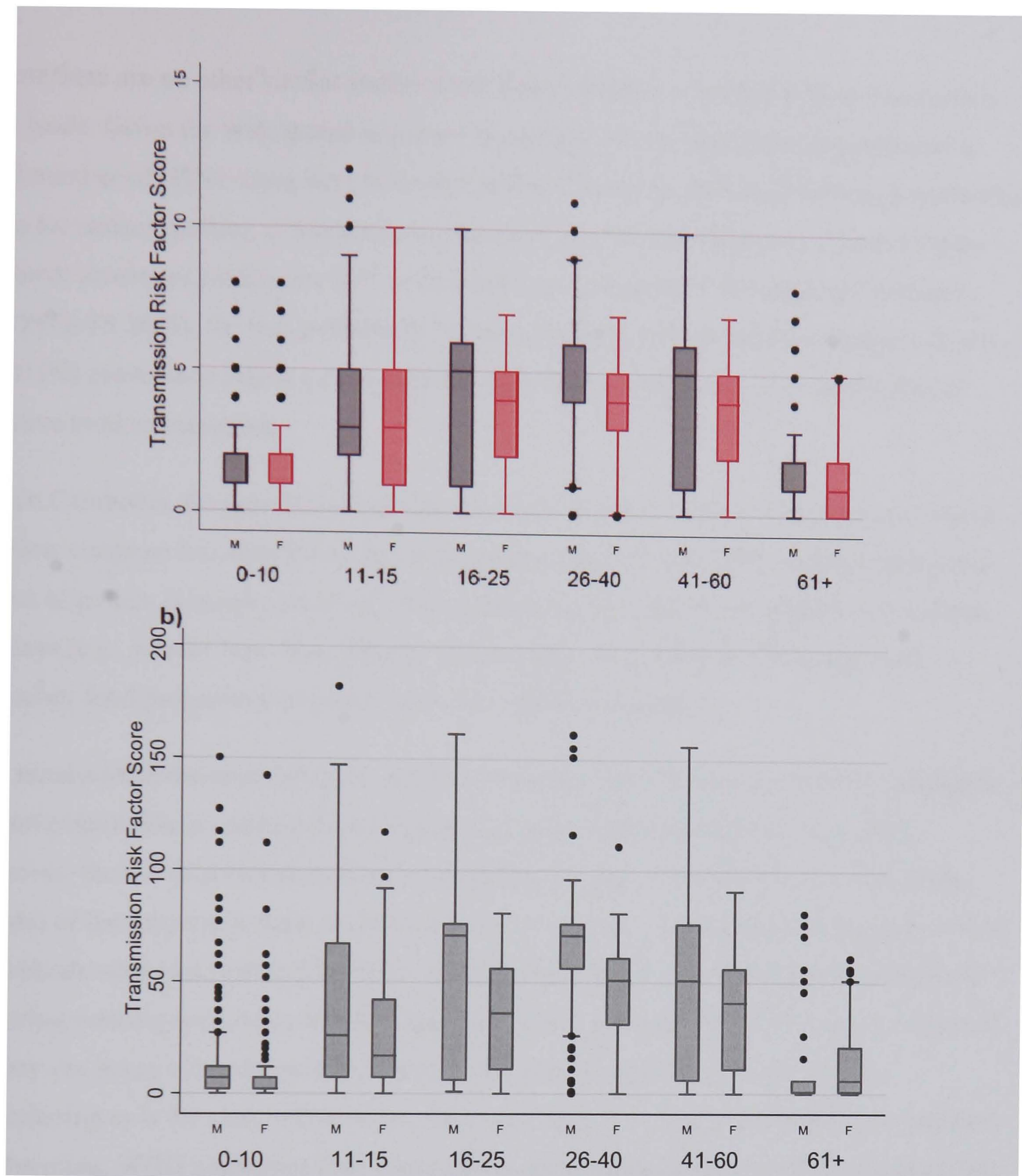


Figure 5-10 (a) Exposure Risk Scores by Age and Gender weighting $\beta_1, \beta_2, \beta_3=1$ and (b) Exposure Risk Scores by Age and Gender weighting $\beta_1=20, \beta_2=5, \beta_3=1$

5.4 Discussion

These results demonstrate that most of the population in rural Cambodia is in frequent contact with domestic poultry, with an estimated 52% of the population carrying out on a regular basis at least one of the practices that one might consider high risk of effective transmission if the bird is infected. I also found that the frequency of exposure to poultry was higher in this study population than that reported in the control subjects used in the Vietnamese (Dinh et al. 2006) and Thai (Areechokchai et al. 2006) case-control studies, suggesting that contact patterns in Cambodia may differ to those in these neighbouring countries. However, at

present there are no other similar studies from these countries to enable a direct comparison to be made. Given the widespread exposure to poultry, it is perhaps surprising that only a small number of H5N1 cases have been reported in Cambodia. Although there is considerable scope for under-reporting of human cases the small number may be due to several factors—the lower density of poultry per km² in Cambodia as compared to Thailand and Vietnam (FAOSTATS 2008), the low probability of people dealing with an infected domestic bird (i.e., low H5N1 prevalence and/or a short duration of infectiousness), and a low probability of effective viral transmission.

Within Cambodia, the typical diet consists primarily of white rice and fish products; animal products compose less than 8% of the daily energy supply (FAO 1999). Eating poultry as a source of protein is usually reserved for special occasions, typically weddings and national holidays (e.g., Khmer New Year [April], Chinese New Year [January/February]) and frequency food preparation of poultry therefore differs seasonally.

It is assumed that the probability of risk from preparing and consuming poultry is negligible if food preparation is conducted under strict hygienic conditions (Greiner et al. 2007).

However, the use of personal protective equipment (i.e., gloves, rubber boots, face masks, aprons) of the subjects in these study areas when in contact with poultry was negligible. Few individuals were observed to have these items in their homes with less than 5% of subjects reporting wearing such items when handling poultry. Inactivation of H5N1 on the surface of poultry can occur when the animal is boiled, therefore if poultry are boiled before defeathering as is the case in Cambodia, the risk of exposure during defeathering is reduced. Furthermore, WHO guidelines state that cooking above temperatures of 70°C will inactivate H5N1 in meats and organs therefore boiling before defeathering would also reduce the exposure potential of individuals cutting/washing meat or internal organs (WHO & INFOSAN 2005).

Although awareness of AI was high among the study subjects, understanding of bird flu was low, especially among children, and at risk poultry handling behaviour continues to be common in rural areas. Despite more than 94% of children 15 years old and younger saying that they have heard of “bird flu,” almost half of them do not believe they are at risk of infection from H5N1. Public awareness campaigns and risk behaviour modification intervention programs should therefore be targeted accordingly.

Even though poultry contact was widespread, there was substantial variation in the frequency of different practices, which although they differed in magnitude according to practice, provide evidence that the potential risk of transmission of H5N1 from poultry to humans is not uniform across age and gender even amongst populations living in close proximity to poultry. Males between the ages of 26-40 reported practices of contact with poultry that give rise to the highest H5N1 transmission risk potential, followed closely by males between the ages of 16-25. This population group differs from the age and sex distribution of the 387 confirmed H5N1 human cases that occurred up to 30 December 2008, in which an excess of cases were observed in children and no differences observed between genders (Writing Committee of the Second World Health Organization Consultation on Clinical Aspects of Human Infection with Avian Influenza 2008). However the group with highest exposure in our study is more similar to the age/sex distribution of the confirmed Thai cases (n=25). The mean age of cases was 22 years and 64% of cases were male (WHO 2006-2009). Such socio-demographic differences in human cases of H5N1 may be because contact patterns with poultry differ between countries. It is also possible that there may be differences by age in intrinsic immunologic susceptibility to infection, pre-existing immunity against human influenza A virus and/or clinical presentation of disease.

This semi-quantitative risk assessment has several limitations and lacks the power of a formal quantitative risk assessment because of epidemiologic data gaps and uncertainties of H5N1 pathogenesis in the host species. To improve future assessments a number of areas would need to be strengthened. First, data are urgently needed on the prevalence of H5N1 in poultry species in regions where H5N1 is recurrent or endemic in domestic poultry flocks. These data are likely to be influenced by the use of biosecurity measures used on farms and in backyard farming settings. While H5N1 poultry outbreaks in countries are reported, because infection may remain asymptomatic in some host species (e.g. ducks), it is difficult to infer prevalence from poultry outbreak reports alone. Prevalence estimates in poultry will allow a greater understanding of the probability that a farm or animal is infected with HPAI/H5N1 (*P*, Figure 5-1).

Secondly, improved knowledge is needed on all the potential routes of transmission of H5N1 from poultry-to-humans and the prevalence of such practices in human populations. I have evaluated what I believe are the main potential routes in which people can become infected with H5N1, however we currently lack sufficient data from the confirmed H5N1 cases

around the world to fully evaluate other potential risk factors for infection such as what role water and other environmental factors play in transmission. Transmission could also include oral ingestion, conjunctival or intranasal inoculation from contaminated water while drinking, swimming or bathing or from faeces while caring for poultry (Vong et al. 2009) and may explain why more children than adults are infected. Furthermore, asymptomatic cases may occur because of low concentrations of viruses in the environment.

Thirdly, an understanding of the influence of genetic and/or immunological factors on transmission is urgently needed since there has been limited yet inefficient human-to-human transmission. There have been several suspected clusters of H5N1 among blood relatives in Indonesia and other countries, however these have been rare occurrences to date (Kandun et al. 2008).

Lastly, virus transmission potential should not be treated as equal across contact practices. Empirical data are needed on virus survival in poultry during food preparation practices, in poultry waste (i.e., poultry scrap, faeces), in soil and in water under different environmental conditions. In addition, data—either experimentally produced or collected during field investigations—are urgently needed on the persistence of H5N1 in poultry tissues. Specifically, which organ, tissue or secretion, if any, has the greatest potential for poultry-to-human transmission. One way of estimating this is to quantify the viral concentrations in various tissues under a variety of conditions (e.g., days post infection, whether or not the animal is exhibiting symptoms, by vaccination status, etc).

Collaboration between human and animal health sectors is essential to understand the risk of transmission between domestic poultry and humans. Current exposure estimates are too general to explain the current pattern or to predict future cases of H5N1 infection in human populations. Rapid, systematic and standardized collection of detailed information on poultry contact patterns in suspected human outbreaks of H5N1 would improve our understanding of transmission from poultry to humans.

Chapter 6 Poultry Contact Networks in Cambodia: Implications for Improving Highly Pathogenic Avian Influenza (HPAI/H5N1) Surveillance and Control

6.1 Introduction

In past epidemics of infectious disease, the movement of humans and animals has been instrumental in the transmission of the disease over great distances. Beginning around 1350AD and for the next 300 years, the transmission of the “Black Death” throughout Europe and Asia can be traced to the movements of humans on foot, horseback and boat (Scott & Duncan 2005). More recently, the worldwide epidemic of SARS illustrated our global connectedness via air travel (Hollingsworth et al. 2007; Hufnagel et al. 2004).

The movement of animals has played a key role in disease transmission as was seen in the 2001 Foot and Mouth Disease epidemics in the United Kingdom (Chis Ster & Ferguson 2007; Ferguson et al. 2001; Ortiz-Pelaez et al. 2006) and HPAI outbreak in the Netherlands (Boender et al. 2007; Stegeman et al. 2004). An understanding of human and animal movement and their contact structures—that is the links between premises via people, animals and equipment—can be used to design more targeted surveillance activities and inform models of disease spread which could result in more cost-effective disease prevention and control (Colizza V et al. 2007; Dent et al. 2008; Green et al. 2008; Hollingsworth et al. 2007; Hufnagel et al. 2004; Kiss et al. 2008; Truscott et al. 2007). Such targeted surveillance are particularly useful in resource limited settings (Stark et al. 2006).

Live bird markets are an important reservoir for HPAI (Woo et al. 2006), as seen in previous outbreaks in the US, Vietnam and Hong Kong (Hayden & Croisier 2005; Horimoto & Kawaoka 2001; Kung et al. 2003a; Nguyen et al. 2005; Wang et al. 2006) and as such, the movement of poultry through markets is potentially important in the circulation and spread of HPAI(Kung et al. 2007; Sims et al. 2003).

Within many countries in Asia, including China, Thailand, Vietnam and Cambodia, some HPAI/H5N1 surveillance programs have focused on live bird markets in which samples are taken and tested for influenza A viruses and/or antibodies on a routine basis. These activities

have shown that H5N1 is circulating in live bird markets (Amonsin et al. 2008; MAFF; Nguyen et al. 2005; Wang et al. 2006). Research has suggested, however, that implementing rest days, in which poultry stalls are emptied, cleaned and restocked can reduce transmission of HPAI and other viruses among birds in live markets (Kung et al. 2003a). Despite their likely role in the circulation and spread of HPAI in South East Asia, little is understood about the poultry market chains or the types and frequencies of contact that exist between rural people raising poultry, local markets and large-national poultry markets in the major cities.

A second cross-sectional survey of rural, peri-urban and urban market sellers and middlemen (i.e., poultry traders) was conducted in Phnom Penh and the same six Provinces (Kampong Cham, Prey Veng, Svay Rieng, Takeo, Pursat, Banteay Meanchey) to evaluate their weekly trading practices and assess seasonal changes in trading and/or poultry movement. The overall aim of the second study is to describe the current movements of poultry throughout Cambodia and examine how these movements influence the potential spread of HPAI at local, regional and national levels.

In addition, the results of this study are used to inform the Cambodia's HPAI surveillance strategies. Within Cambodia, the financial and personnel resources of the veterinary services have been limited and unable to implement nationwide continuous surveillance and/or vaccination programs. Risk based surveillance recommendations take into account financial and human resources constraints by the veterinary infrastructure (Snow et al. 2007; Stark et al. 2006).

Three months before data collection ended for this study (September 2007), NaVRI in collaboration with FAO Cambodia began to survey ducks in 14 markets located in 6 Provinces, increasing to 24 markets in 11 Provinces. Their aim was to detect the "presence of HPAI/H5N1 in the major duck producing regions of Cambodia, to estimate the level of risk that ducks pose to poultry (chickens and ducks in traditional and commercial enterprises, to enhance the public awareness of Avian Influenza in poultry markets and to reduce the risks of transmission from ducks to poultry and humans by greater understanding of the epidemiology of HPAI in ducks and the development of effective control measures" (MAFF Unpublished Data).

6.1.1 Objectives of the Chapter

The objectives of this chapter are:

- Identify all poultry markets within Phnom Penh and the 6 study areas;
- Describe and map basic selling characteristics of markets;
- Identify middlemen responsible for transporting live poultry in the 6 provinces and into and out of Phnom Penh;
- Determine the extent and frequency of occupational poultry handling behaviour of study subjects;
- Characterize and illustrate poultry trading practices of rural Cambodians, market sellers and middlemen;
- Characterize the potential role of networks in HPAI/H5N1 virus circulation; and
- Highlight where interventions need to be targeted in the case of poultry outbreaks or if possible where vaccination should take place.

6.2 Methods and Materials

6.2.1 Data Collection: Rural Cambodians

Poultry movement and trading data from rural Cambodians was collected during the first cross-sectional survey of rural Cambodians in six provinces (Kampong Cham, Prey Veng, Svay Rieng, Takeo, Pursat, Banteay Meanchey) from November – December 2006 and November-December 2007 as described in Chapter 3. The survey included 115 village chiefs (1 per village) and 600 heads of households (100 per province).

In summary, standardized questionnaires were administered by trained interviewers in Khmer. In addition to the topics of questions described in Chapter 3, additional questions were included in the questionnaires for the heads of household and village chiefs to collect data on poultry trading practices from within and outside of their home village. Questionnaires for the Head of Household and village chiefs are provided in Appendix B. The additional questions on poultry trading practices are described below:

Village Chiefs: Variables included the frequency of poultry trading via middlemen from their village and destination where the poultry were sold.

Heads of Household: Variables included the frequency of selling poultry within and outside of their village, the destination where poultry were sold, the quantity of poultry sold (by species) to each destination, and the use of middlemen for trading poultry.

6.2.2 Data Collection: Poultry Traders

6.2.2.1 Identification of poultry traders and markets

In rural areas of the above provinces and in Phnom Penh, meetings were held with provincial and district level veterinarians to identify all markets located within the study districts that sold poultry. Visits were made to each named market and discussions were held with local market sellers to determine 1) if poultry were sold at the markets 2) the time of day poultry sellers sold at the market, 3) if middlemen worked at the market and if so, 4) when and where they were likely to frequent the market. For the purposes of this study, I defined an eligible poultry market as those that contain ≥ 3 poultry selling stalls. Return visits were made to all eligible markets to interview all available market sellers and middlemen.

Snowball sampling methods (Wasserman & Faust 1994) were utilized to identify poultry selling markets, poultry sellers and middlemen responsible for trading poultry in the study areas and into Phnom Penh. Snowball sampling techniques are used when the quantity of individuals in the sample (e.g., poultry markets, market sellers and middlemen) are unknown. There are no known lists of markets, market sellers or poultry traders available from the Ministry of Agriculture, Fisheries and Forestry or any other source. To obtain information on poultry markets in Phnom Penh and the trading of poultry into Phnom Penh, a focus group discussion was conducted with Phnom Penh market veterinary inspectors. Twenty-one vet inspectors from 15 markets were split into two groups and both discussed the two main objectives, which were to identify all markets selling poultry within Phnom Penh and gain insight into how to identify middlemen responsible for trading poultry at these markets (Figure 6-1). Market lists prepared by the two groups were compared and a final list of markets was created.



Figure 6-1 Focus group discussions with Phnom Penh market vet inspectors

Permission was obtained from all subjects to take and use photographs in reports.

All markets identified through the focus group discussions were visited by field staff to collect basic data on the market selling characteristics, including the animals sold at the market (chickens, ducks, geese, cattle, pigs, fish, other), the number of live and “prepared” (i.e., boiled and defeathered at home prior to sale at the market) poultry stalls, and if poultry sold at the market were caged, tied together or free-ranging.

Veterinary inspectors who formed the focus-groups were not able to identify middlemen trading poultry with Phnom Penh markets, but provided contact details of members of the Office of Animal Health and Production in Phnom Penh. The Office of Animal Health and Production in Phnom Penh holds similar responsibilities for animal health as NaVRI except that they are responsible for animal health in Phnom Penh whereas NaVRI is responsible for animal health throughout the entire country. In-depth face-to-face semi-structured interviews were held with the Chief of Animal Movement in Phnom Penh and his staff who provided information about animal movement into the capital. During these interviews, information was obtained on poultry movements into Phnom Penh, the number of middlemen thought to be responsible for trading poultry in the six study provinces as well as into Phnom Penh and the transport patterns of middlemen, which were used to determine the most likely locations for interviews with middlemen.

Following interviews, field visits were made to veterinary inspection points located along all six national roads leading into Phnom Penh and three poultry markets in Phnom Penh identified as the “main poultry selling markets” (i.e., Orussey, Chba Ampov and Deum Kor Markets). As a result of my interviews with vet inspectors from these three markets and middlemen, approximately one dozen poultry stock houses located near these three main poultry selling market in Phnom Penh and 16 semi-commercial poultry farms were identified as being integral in the movement patterns of poultry. Field visits were also made to each of these locations.

6.2.2.2 Interviews of market sellers and middlemen

Structured interviews with middlemen took place during field visits to markets which coincided with the known and suspected times the middlemen traded at each market, inspection points along national roads, stock houses in Phnom Penh and semi-commercial farms in Phnom Penh between October 2006-April 2007 and October-December 2007 (Figure 6-2). Structured interviews of market sellers took place during field visits at the time

of day when market sellers were known to sell poultry (identified by district veterinarians and discussions with market sellers) and included all available market sellers at the markets. Repeat visits to the three main markets in Phnom Penh were made until one market seller per stall was interviewed.



Figure 6-2 Interview of middlemen at inspection point along National Road No. 6

Permission was obtained from all subjects to take and use photographs in reports.

The questionnaires for the market sellers and middlemen are provided in Appendix F. The variables included in each questionnaire are described below.

6.2.2.3 Questionnaires for poultry market sellers

Questionnaires were designed for poultry market sellers to address:

Market Level Questions: The first page of the poultry market seller questionnaire was observational and aimed to collect information at the market level and the stall level. Variables included: GPS coordinates of the market; which animals were sold at the market (yes/no; chickens, geese, singing birds, fighting cocks, ducks, pigs, cattle, fish, other); how each species was sold at the market (observed; alive, dead, sold whole or in part and whether organs of the animal are sold); the number of stalls at the market that sold live poultry and prepared (slaughtered away from the market) poultry (observed and supplemented by asking other market sellers); characteristics of how the birds were kept at the market (observed;

free-ranging, caged, tied together); and if poultry faeces were visible on the ground of the market (observed; yes/no).

Stall Level Questions: With the exception of how many poultry stalls were at the market, the same set of observations that addressed characteristics of the market were used to address the characteristics of the market sellers poultry stall.

Demographic Information of the Market Seller: Variables included: age (years old) and gender of the market seller; number of years they had traded at the market; and what type of trade was conducted at the market (bought only, sold only, bought and sold). Permission was sought to contact the market seller again and a mobile phone number was recorded if they owned a mobile phone.

Poultry Selling Characteristics: Variables addressed to the market seller included: number of people by gender that were working at the stall on the day of interview; quantity of people (by gender) responsible for preparing poultry for sale; quantity of people (by gender) responsible for boiling, bleeding, defeathering, removing internal organs, butchering of their poultry; the location (home or at market) where boiling, bleeding, defeathering, removing internal organs, butchering of their poultry took place; the use of gloves, boots, aprons, face masks (plastic or cotton) or other PPE on the day of interview (observed and recorded as yes/no); the number of times per week they cleaned their poultry cages and selling areas; and the use of disinfectant when cleaning their stall (yes/no) (Figure 6-3).



Figure 6-3 Market seller from Poipet (Akak) Market, Banteay Meanchey province, Cambodia

Permission was obtained from all subjects to take and use photographs in reports. Note the cage for live poultry, poultry scrap pile and resting quarters behind the selling platform.

Use of middlemen in poultry trade: Variables included: number of days per week live chickens and ducks were received from middlemen; quantity of animals they received per shipment (if the subject was not able to provide an absolute quantity, a range of animals was recorded); and method of transportation of their poultry shipments (motorbike, car, truck, other).

Origin of poultry: Variables included: number of middlemen they purchased poultry from (interviewers recorded “0” if market sellers did not use middlemen and purchased poultry themselves); and the origin of the poultry they sold (country, province, district, village and/or market).

Seasonality of trading: Market sellers were asked to list periods of increased trading (e.g., holidays or festivals). Variables included: the name of the holiday or festival that was responsible for increased trading; and quantity of chickens and ducks sold in the 4 weeks prior to the listed holiday (if the subject was not able to provide an absolute quantity, a range of animals was recorded).

6.2.2.4 Questionnaires for middlemen

Questionnaires were designed for poultry market sellers (Appendix F) to address:

Demographic information of subject: Variables included: age (years old), gender, number of years employed as middleman; and the number of villages visited each day (#) and week (#) to purchase poultry. Permission was asked to contact the market seller again and a mobile phone number was recorded if they owned a mobile phone. In addition, GPS coordinates were taken at the location of the interview.

Origin of poultry: Variables included: the location(s) of origin where poultry was purchased (country, province, district, village and/or market); the number of visits each week to the named location(s); the quantity of chickens and ducks purchased at each location; the state of the poultry at the time of purchase (alive, prepared [i.e., dead: boiled, defeathered, internal organs removed] or dead and not prepared).

Destination poultry were sold: Variables included: the number of locations to which they sold poultry; the name of the locations(s) where poultry were sold (i.e., destination: country, province, district, village and/or market); the estimated distance between the origin and destination of each poultry trade (km); the quantity of chickens and/or ducks sold to each destination each week. Middlemen were also asked if they purchased poultry from or sold poultry to Vietnam (yes/no) and Thailand (yes/no); and if they purchased dead or sick animals (yes/no).

Poultry Transport: Variables included: mode of transportation used for chickens and/or ducks (moto, car, truck, other specified); if chickens and/or ducks were transported alive, dead or both alive and dead; if chickens and ducks were mixed during transport (yes/no); the quantity of chickens and/or ducks carried on their vehicle; the use of cages during transport (yes/no). If cages were used, middlemen were asked what the cages were made of (wood, plastic, or metal); if cages were stacked on their vehicle (yes/no); if there were trays to catch faecal matter beneath each cage (yes/no); and if cages were cleaned after each transport (yes/no).

Selling Practices: Variables included: if all poultry traded by the middleman each day was sold on the same day (always, sometimes, never); and what they did with poultry that they were unable to sell during the day (bring home, send them to slaughter, bring to other markets, bring back to other farms or other).

Seasonality of Trading: Middlemen were asked to list periods of increased trading (e.g., holidays or festivals). Variables included: the name of the holiday or festival that was responsible for increased trading; quantity of chickens and ducks sold in the 4 weeks prior to the listed holiday (if the subject was not able to provide an absolute quantity, a range of animals was recorded).

Illustration of Movement via Middlemen: At the end of each questionnaire, interviewers were instructed to sketch the path of poultry from origin to final destination. This was included in the questionnaire as a method to verify that the data collected via the questions would represent the actual path of movement from the point of origin to the point of destination. An example of this is shown in Figure 6-4 below.

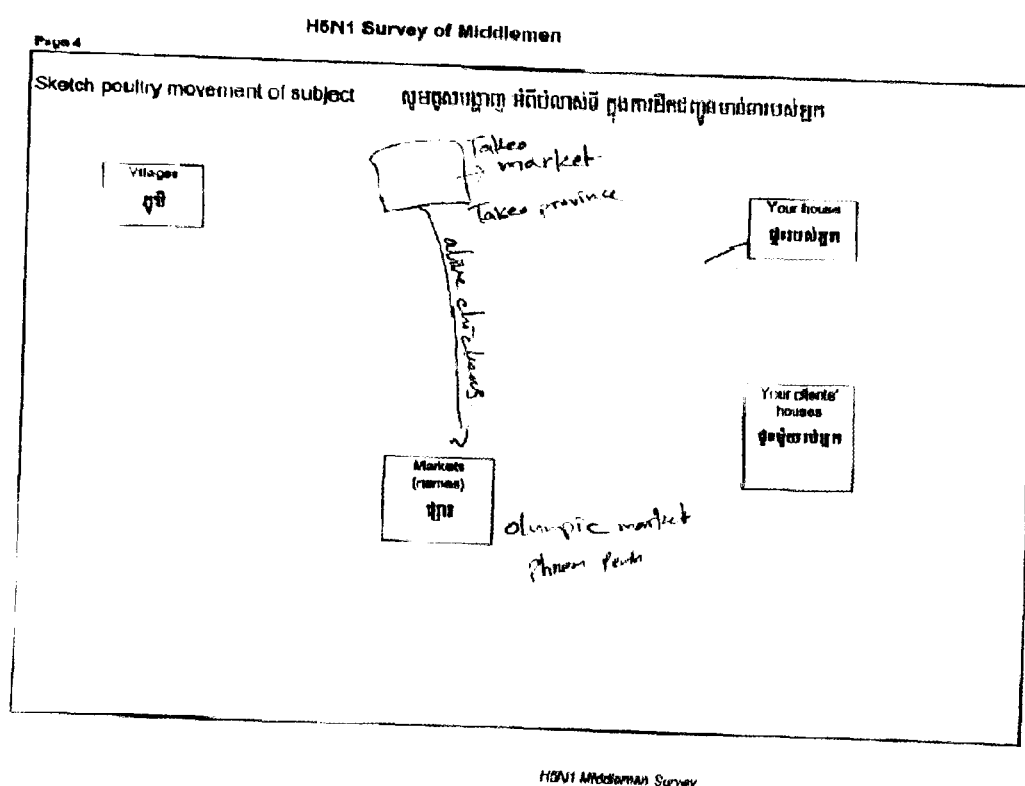


Figure 6-4 Example of a sketch of poultry movement collected from a study subject

This illustration provides an example of how the movement of live chickens from Takeo Market, Takeo Province to Olympic Market in Phnom Penh was captured in the questionnaire.

6.2.3 Recruitment of Staff, Interviewer Training and Questionnaire Piloting

Three interviewers were trained to administer the questionnaires in Khmer. Piloting of the questionnaires took place in rural poultry selling markets to determine the best approach of administering the questionnaire so as to minimize disruption of selling and how to structure the questions to best obtain origin of purchase and final destination where the poultry were sold. We encountered difficulties in identifying the appropriate time to administer the questionnaire since poultry sellers were very busy while at the markets. I found that the best approach was to be at the markets before they began selling and remain in the market until

after they finished selling their poultry to wait for moments where they were free to spend a few minutes to be interviewed.

The questionnaires were modified to improve the nature in which poultry origin and destination information was recorded. I found that it was best to include a table format (Figure 6-5) to capture these data.

ID Code: | _____ |

H5N1 Survey of Middlemen

Page 2

1. How long have you worked as a middleman transporting poultry? _____ years

2a. How many villages do you visit each day? _____ villages/day >>>>> 2b. How many villages do you visit each week? _____ villages/week

3 Where are the villages located where you **BUY** chickens and ducks?

	Province	District	Village	# visits each week	Quantity purchased each week?		Are animals (1) alive, (2) prepared, (3) dead, not prepared when purchased?		
					Chickens	Ducks	1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP
3a	<input type="checkbox"/> Vietnam						1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP
3b	<input type="checkbox"/> Thailand						1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP
3c	<input type="checkbox"/> Cambodia #1						1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP
3d	<input type="checkbox"/> Cambodia #2						1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP
3e	<input type="checkbox"/> Cambodia #3						1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP
3f	<input type="checkbox"/> Cambodia #4						1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP
3g	<input type="checkbox"/> Cambodia #5						1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP
3h	<input type="checkbox"/> Cambodia #6						1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP
3i	<input type="checkbox"/> Cambodia #7						1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP
3j	<input type="checkbox"/> Cambodia #8						1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP
3k	<input type="checkbox"/> Cambodia #9						1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP
3l	<input type="checkbox"/> Cambodia #10						1 <input type="checkbox"/> A	2 <input type="checkbox"/> P	3 <input type="checkbox"/> D/NP

Figure 6-5 Table in middlemen questionnaire to obtain poultry origin data

6.2.4 Data Entry and Analyses

All data were entered into EpiData v 3.2 and analyzed using STATA v10 (StataCorp, College Station, Texas).

The study population is described as the number of subjects recruited and interviewed for each subject type (head of household, village chief, market seller and middleman). Gender is presented as the number and percent male for each subject type. Descriptive data on age is presented as median and IQR and is presented for both male and female subjects. Age distributions were compared using Wilcoxon rank sum tests for males and females separately. Binary questions of poultry selling characteristic are presented as numbers and percentages and compared across geographic regions. Associations between categorical variables were tested by cross-tabulating them in a contingency table and chi-square tests or Fishers exact tests were used for statistical tests of significance as appropriate.

The use of personal protective equipment by market sellers was analyzed by gender and geographic region using chi-squared or Fisher's exact tests, as appropriate.

Reported distances from the point of origin where poultry were purchased to the destination where poultry sold via middlemen were summarized using the median and IQR. Distance was also aggregated into 50 km categories (e.g., 0 [traded within the same district], 1-50, 50-99, 100-149, 150-199, 200-249 and 250+ km) and the median and IQR of the number of poultry transported for each journey was calculated.

6.2.4.1 Transforming data on quantity of poultry trade

During the data collection, my interviewers found that some subjects felt it was easier to quantify the amount of birds sold in kilograms (kg) rather than the number of birds. Any data on quantity of poultry that was collected as weight (kg) sold per week was converted to number of birds using the average weight of chickens and ducks sold at Orussey Market in April 2008 (average weight of chickens was 1.2 kg/chicken; 1.8 kg/duck) (personal communication with the Office of Animal Health and Production, Phnom Penh). For data on the quantity of poultry that was reported as a range, I felt that the minimum value provided by subjects was a more accurate estimate of the quantity and thus the minimum value was used in the analyses among those that provided a range.

6.2.5 Contact Network Analyses and Interpretation

Social network methods were used to characterize the network of poultry movements in the study areas (Scott 2000; Wasserman & Faust 1994). Data on the origin of purchase, destination of sale, and weekly quantities of chickens and ducks traded between the two locations were used to create a directed network of chicken and duck movements by market sellers and middlemen within and into Cambodia.

The locations where poultry were purchased and sold for each subject were reformatted into source-destination pairs that included the quantity of poultry (chickens and ducks) traded each week across each source-destination pairing using STATA v10 (StataCorp, College Station, Texas). Poultry quantity data was aggregated for each identical source-destination pair. Poultry network adjacency matrices (Figure 6-6) were developed separately for chicken and duck movement.

Source	Destination									
	A'	B'	C'	D'	E'	F'	G'	H'	I'	J'
A	0	0	0	0	0	0	0	0	0	0
B	60	0	0	0	0	0	0	0	0	0
C	0	0	0	0	115	0	0	0	0	0
D	0	0	0	0	0	0	0	0	0	0
E	0	0	0	0	0	0	0	0	0	0
F	0	156	0	0	0	20	0	0	0	0
G	0	0	0	30	0	500	0	0	0	0
H	0	0	0	0	0	0	0	0	0	0
I	0	0	0	0	0	0	0	0	0	0
J	0	0	0	0	0	0	0	0	0	0
K	0	0	0	0	0	0	0	0	0	0
L	0	0	0	0	0	0	0	0	0	0
M	0	0	0	0	0	0	0	0	0	0
N	0	0	0	0	34	0	0	0	0	0
O	0	0	0	0	0	0	0	0	0	1000
P	0	0	0	0	0	0	0	0	0	0
Q	0	0	0	0	0	0	0	0	0	0
R	0	0	0	0	0	0	0	0	0	0
S	0	0	0	0	0	0	0	0	0	0

Figure 6-6 Example of an adjacency matrix

The letters A-S in the first column represent the source and A'-J' in the first row represent the destination, where A=A', B=B' etc. The value in the cells between the source and destination (e.g., F=B') indicated the quantity of animals (chickens and ducks, e.g., 156) that are traded between the source and destination on a weekly basis. The chicken and duck adjacency matrices were composed of 175 X 175 locations.

6.2.5.1 Visualization of the networks

The networks were illustrated using NetDraw v2.055 (Borgatti 2002). In the resulting networks, nodes indicate locations and were weighted using in- and out-degree. The colours of the nodes indicate location type (e.g., market, stock house, rural farm or household, commercial farm, semi-commercial farm, foreign source). Edges or ties linking nodes illustrate the direction of poultry movement as indicated by arrows and tie strength is indicated by the thickness of arrows (i.e., the thicker the arrow, the more poultry passing between from the two points).

In Figure 6-7 below, which is an illustration showing an example of a directed network, the dots represent nodes (i.e., locations) and the links are represented by directional arrows.

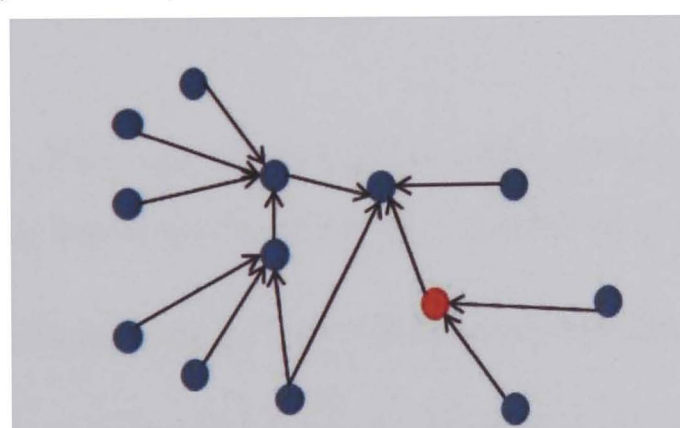


Figure 6-7 Illustration of an example of a directed network

The dots in this figure represent nodes and ties are directed as shown by arrows

6.2.5.2 Characterization of a network

The group and sub-group structure of the networks were explored by examining the number of components (i.e., the number of connected groups) within each trade network; the core and periphery, which is a way to measure areas of low and high cohesion in the network; the cut-points, which if removed would divide the network; and centralization, which is the extent to which the network revolves around a single node (Figure 6-8) (Scott 2000; Wasserman & Faust 1994).

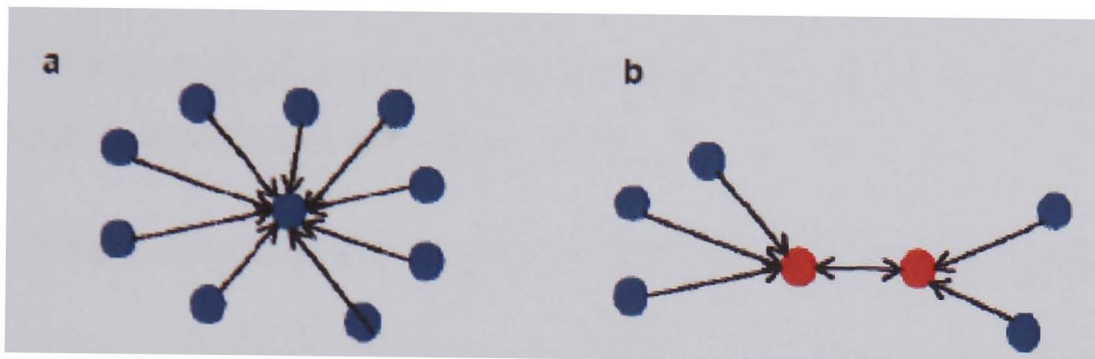


Figure 6-8 Examples of (a) a highly centralized network and (b) a network with clear cut points

In figure b, the nodes in red, if removed would clearly divide the network in half.

As a measure of how well connected the nodes (locations) are within the network I calculated the in-degree (quantity of birds terminating at the node) and out-degree (quantity of birds originating from the node) for each network using UCInet software (Borgatti et al. 2002). Degree is the number of nodes adjacent to a given node, however in directed network degree is measured as in-degree, i.e., the number of links terminating at the node; and out-degree, i.e., the number of links that originate from the node (Scott 2000; Wasserman & Faust 1994). In my analyses, in-degree measures the quantity of birds terminating at the node and out-degree measures the quantity of birds originating from the node. In Figure 6-8b, the red node on the right hand side of the diagram has an in-degree of 2 and an out-degree of 1. Degree is calculated as $\sum_j a_{ij}$ where a = adjacency matrix.

Betweenness is a measure of the number of shortest paths between other points in the network that a node lies on and is a measure of the importance of the node in connecting

other nodes (Scott 2000; Wasserman & Faust 1994). Betweenness is calculated as $\sum_{i,j} \frac{g_{ijk}}{g_{ij}}$

where g_{ijk} is the number of geodesics (i.e., shortest path between nodes i and j) including the node of interest (k) and g_{ij} is the total number of geodesics between i and j .

6.2.6 Risk-Based HPAI Surveillance Recommendations

Targeted surveillance recommendations were developed for the National Veterinary Research Institute (NaVRI) to improve their active HPAI surveillance activities. Recommendations were developed to identify locations in which early detection of infection is most likely to prevent transmission from poultry-to-humans and interrupt poultry-to-poultry transmission. The recommendations developed for NaVRI were based on the following objectives of the surveillance:

1. Objective 1: Monitor the HPAI status of poultry populations in rural areas
 - Nodes with the highest in-degree were enumerated as those most likely to detect HPAI if it were present in the market chain.
2. Objective 2: Early detection of incursion in nodes (markets) with high potential for spread
 - Nodes that are most likely to interrupt poultry-to-poultry transmission are those with greatest number of connections and the highest out-degree scores.

6.2.6.1 Connectedness of the networks: evaluation of the potential for spread of H5N1

In addition, I also explored the potential spread of H5N1 outbreak across the network by seeding a hypothetical H5N1 outbreak at various premises in the networks and examining how a disease might spread across a network via links of poultry trade. Figure 6-9 below is an example of how a disease might spread across a network via links of poultry trade. In this example, the virus was seeded in the node identified by the black circle. The gray nodes indicate the potential spread of the disease via directed trade links between the nodes.

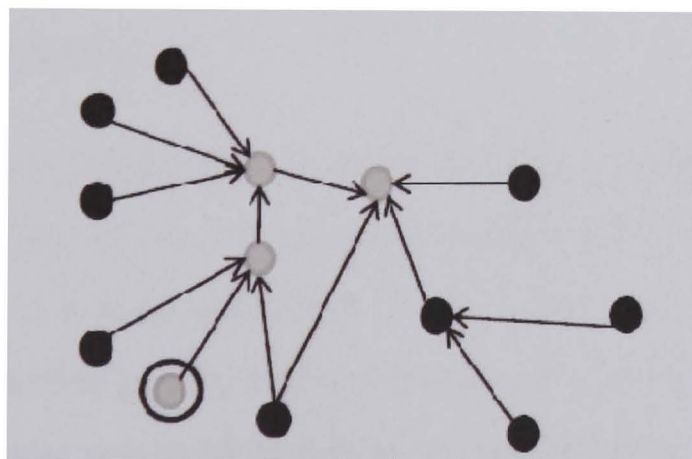


Figure 6-9 Example of disease spread across duck network

Node colour indicates infected (grey) or uninfected (black) premises; black circle indicates the premises where H5N1 is hypothetically detected

The undirected links of the networks were viewed as potential infectious links between nodes. Likely routes of transmission included faecal-oral route via viral shedding of infected poultry

and/or environmental contamination in the market by fomites, people and/or viral shedding of birds. However, I assumed that each of the directed links resulted in the effective transmission of the virus via the birds, fomites or people moving between the two nodes. No transmission dynamics were included in this exercise; however I treated the duck and chicken networks separately because of the difference in pathogenicity and presence of symptoms in the two species (for example (Saito et al. 2009)).

This exercise was repeated by seeding a HPAI outbreak in each of the 15 highest out-degree locations (which included locations in Vietnam) and separately if an outbreak were detected in the location with the highest in-degree value (Orussey Market). The total outbreak size of each seeded outbreak was calculated as the total number of nodes connected to the infected premises.

6.2.7 Ethical Considerations

Ethical approval was granted from the Cambodian Ministry of Health and London School of Hygiene and Tropical Medicine ethical committees. Prior to sampling, field visits were conducted and meetings were held with provincial veterinarians and market veterinary inspectors to explain the study objectives and procedures. Written informed consent was obtained from all rural Cambodians. Verbal consent was obtained from all village chiefs, market sellers and middlemen prior to interview.

6.3 Results

6.3.1 Subject and Market Characteristics

6.3.1.1 Poultry market identification

A total of 122 markets were identified in the study districts, of which 102 were identified as markets selling poultry (Figure 6-10), including 43 markets in Phnom Penh, five in Banteay Meanchey, six in Pursat, 11 in Kampong Cham, 15 in Takeo, 15 in Prey Veng and seven in Svay Rieng. An additional nine poultry selling markets were identified by middlemen that were outside of the provinces surveyed including one in Kampong Chhnang, one in Kampong Speu, one in Kampot, two in Kampong Thom, three in Kandal and one in Vietnam. Figure 6-11 shows images of the poultry selling areas of the three main poultry selling markets—Orussey, Deum Kor, and Chba Ampov markets—in Phnom Penh.

The median number of markets identified per district was 3 (IQR [1-4]; n=102); within Phnom Penh the median number of markets per district was 4 (IQR 4-8.5; n=42).

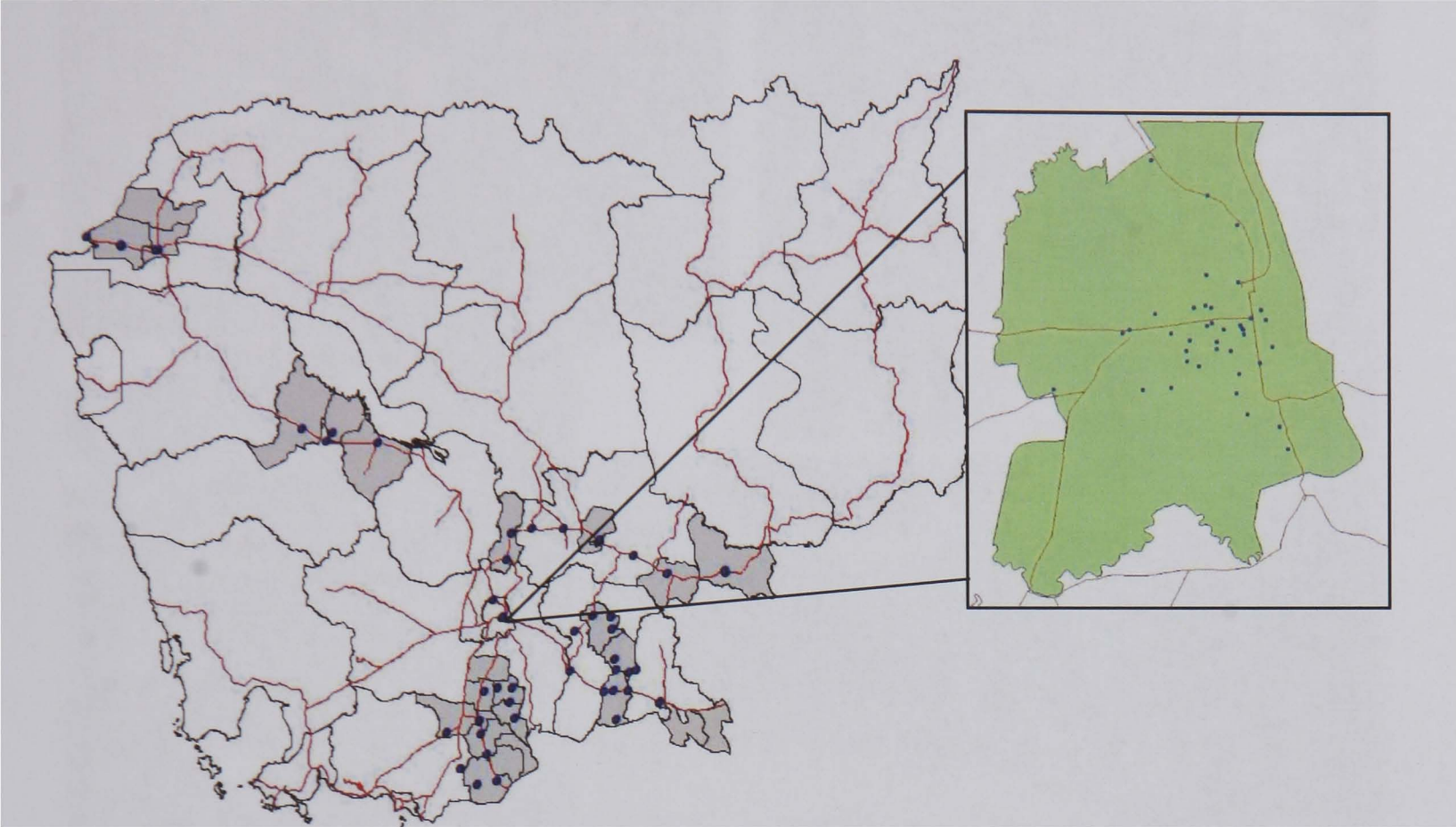


Figure 6-10 Markets locations (blue dots) identified in study districts (shaded gray) in Cambodia; the inset identifies market locations in Phnom Penh



Figure 6-11 Poultry markets in Phnom Penh

Upper left: Orussey Market slaughtering area; upper right and lower left: Deum Kor poultry selling areas; bottom right: live bird selling area of Chba Ampov Market

Detailed selling characteristics were collected from 62 markets. All sold prepared or live chickens and prepared cattle, 64.5% (n=40) sold live or prepared ducks, none sold geese or fighting cocks, and almost all sold prepared pigs (96.8%, n=60) and fish (95.2%, n=59). Among markets that sold live chickens or ducks (n=31/62; 50%), 32.2% (n=10/31) kept live poultry caged together while 67.8% (n=21/31) kept poultry together at the markets.

The median number of stalls selling poultry (live or prepared) at the markets was 5 (IQR 3-7, max 34, n=62). All of the identified markets in Phnom Penh (n=43) sold prepared (boiled and de-feathered) poultry and approximately half (46.3%) sold live poultry and slaughtered poultry on the premises, i.e., are considered live animal wet markets. Approximately 10% (6/62) allowed poultry to be free-ranging in the markets. However there were poultry faeces visible on the ground at the market in 35.5% (n=22) of markets.

6.3.1.2 Subject recruitment

A total of 115 village chiefs and 600 heads of households (100 from each study province) from 115 villages were recruited and interviewed, as were 120 middlemen, and 102 market sellers from the study provinces and in Phnom Penh. There were no refusals to participate in the study by village chiefs, heads of household and market sellers. There were a few (<5) refusals among middlemen because they felt they were too busy to be interviewed; among those, most agreed to provide their province of origin (poultry source) and allowed observational data (e.g., gender, species trading, mode of transport) to be collected. These data were included in the relevant analyses.

Village chiefs, heads of household and middlemen were predominately male, while market sellers were predominately female (84.3%; Table 6-1).

Table 6-1 Characteristics of poultry traders recruited

Characteristic	Middlemen	Market Sellers	Village Chiefs	Heads of Household
N	120	102	115	600
Gender (n [%] male)	78 (65.0)	16 (15.7)	108 (93.9)	378 (63.0)
Age (median, IQR)	35 (29-42)	40 (28-48)	52 (48-58)	46 (37-56)
Home residence (n provinces)	9 (n=111)	8	6	6
Length of occupation years (median, IQR)	10 (5-14)	10 (5-20)	--	--
Villages visited each week (median, IQR)	3 (2-5) ^a	--	--	--
Selling characteristics n (%)				
Sell any poultry	120 (100)	102 (100)	-- ^b	24 (4.0) ^c
Chickens	102 (85.0)	96 (94.1)	--	23 (3.8)
Ducks	29 (24.2)	53 (52.0)	--	3 (0.5)
Chickens & ducks	19 (15.8)	49 (48.0)	--	2 (0.3)

^a n=110; -- not assessed; IQR=Interquartile Range

^b Village Chiefs reported on selling practices of the village rather than self

^c Sell poultry outside of home village

6.3.1.3 Preparation of poultry for sale

The median number of males and females per stall responsible for preparing poultry for sale (boiling, bleeding, defeathering, removing internal organs, butchering at the market) was 2 (IQR: 1-3, range 1-7) and 1 (IQR: 1-2; range 1-5), respectively. Table 6-2 summarizes those responsible for each individual practice involved in preparing poultry for sale at the markets.

Table 6-2 Numbers of people responsible for the practices associated with preparing poultry for sale at markets

Practice	Males median (range)	Females median (range)
Boil poultry	1.5 (1-7)	1 (1-5)
Bleed poultry*	1 (1-7)	1 (1-5)
Defeather poultry	2 (1-7)	1 (1-5)
Remove internal organs	2 (1-7)	1 (1-5)
Butcher (cut meat) poultry	2 (1-7)	1 (1-5)

*Slaughter by cutting the throat and bleeding poultry

6.3.1.4 Use of personal protective equipment by market sellers

The use of personal protective equipment (PPE) by poultry market sellers was minimal or non-existent. The largest number of subjects using any PPE were from markets in Phnom Penh (n=67) where 17.9% of market sellers were observed wearing gloves while handling poultry, 13.4% were wearing face masks, 7.5% were wearing rubber boots and 7.5% were wearing aprons. Only one market seller from outside of Phnom Penh (1/53) was observed wearing any PPE while handling poultry. Eighty-nine percent of market sellers (83/93) reported cleaning their selling stalls at least once per day, however only 18.1% (n=15) reported cleaning with disinfectant.

6.3.2 Characterizing Poultry Movement

6.3.2.1 Poultry traders

Few of the 600 rural Cambodians interviewed reported selling chickens (3.8%) or ducks (0.5%) outside of their home village during the previous eight-month period (Table 6-1). None used middlemen to sell poultry and none were able to provide the locations where poultry were sold. Eighty-one percent of village chiefs (n=142) provided information on the destination of poultry sale via middlemen from their village, but were not able to quantify the number or specify the species of birds traded.

Focus group discussions and in-depth semi-structured interviews with animal health officials suggested that approximately 80-90 middlemen transported poultry into Phnom Penh on a daily basis. However, our field investigations identified and interviewed 120 middlemen. Almost all market sellers were able to provide the origin (i.e., source) of the poultry they traded. Therefore, poultry movement data from village chiefs in the 24 districts sampled, market sellers in the study areas and Phnom Penh and middlemen identified through

interviews with market sellers, vet inspectors and recruited at markets in the 24 districts and Phnom Penh and on the main routes to Phnom Penh were available for use in the network analyses.

Additionally, 16 semi-commercial farms near Phnom Penh, which were identified from vet inspectors and middlemen interviews with holdings of 2,000-4,500 chickens or 400-2,000 ducks (no mixed species farms were identified), were visited and the owners were interviewed. These farms do not practice an all in/all out system. Rather there is a “continuous flow of stock” (Kung et al. 2007) moving through the farm each week. The predominant sources of ducks from these farms were from Vietnam, which are purchased at 1-2 days old and are unvaccinated for H5N1. Farm owners reported that these ducks are reared for approximately two months before selling to markets in Phnom Penh.

Furthermore, approximately one dozen stock houses were identified throughout Phnom Penh from interviews with Phnom Penh veterinarians, market sellers and middlemen. The function of these homes is to collect (purchase) poultry from middlemen trading poultry from provinces outside of Phnom Penh, usually having travelled >50 km, and then sell to markets in Phnom Penh directly. These homes are equipped with large cages used to store live poultry. All stock house owners interviewed reported that poultry were sold to markets in Phnom Penh on the same day of purchase. Thus poultry were not kept in the cages for more than 8 hours before being sold onto markets in Phnom Penh.

6.3.2.2 Distances moved

Poultry is transported by middlemen from nine Cambodian provinces and across the borders from Vietnam and Thailand. The median distance that poultry travelled from its source was 70 km (IQR 15.5-99.5) and the majority (84.3%) of poultry movement via middlemen was >10 km from the source and directed into Phnom Penh. The quantity of poultry transported each week by distance the poultry was traded (distance between the point of purchase and point of sale) is shown in Figure 6-12.

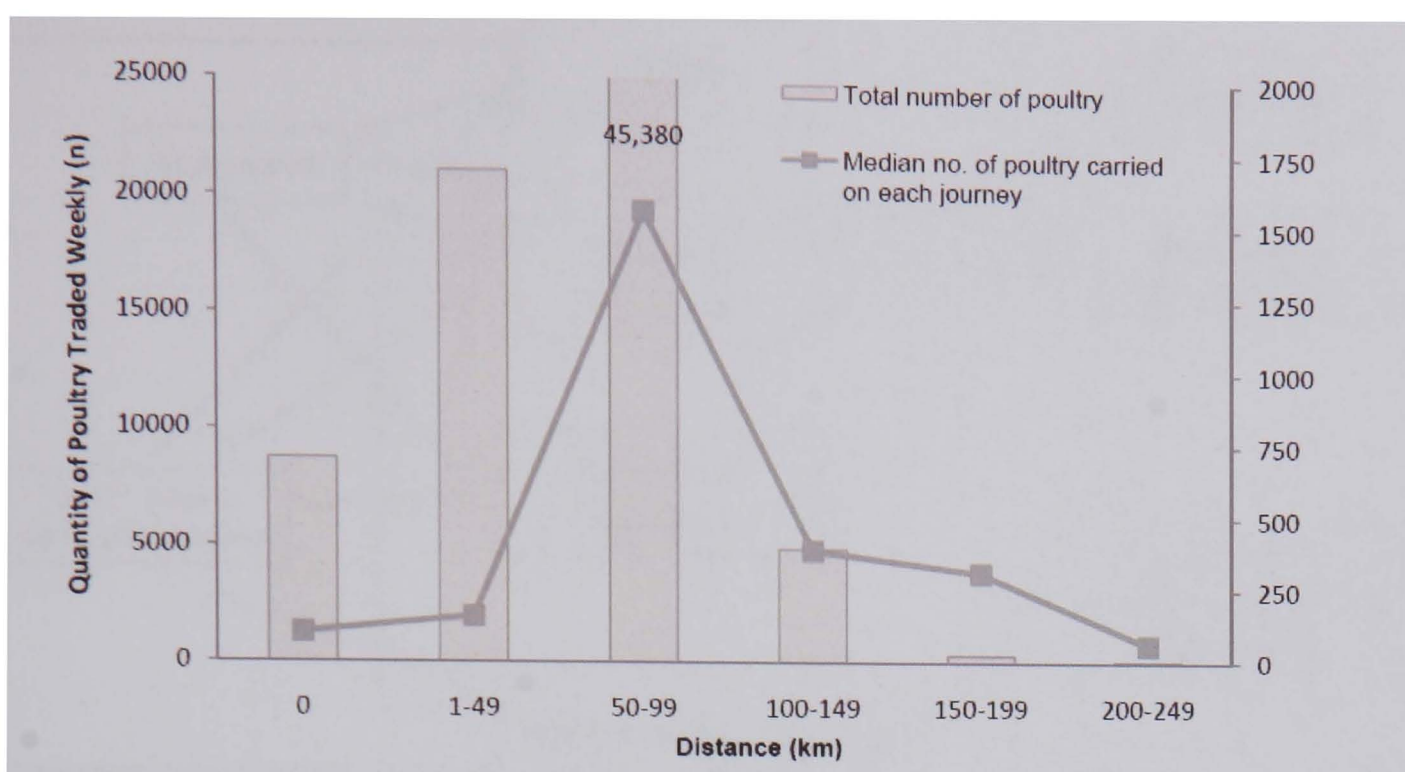


Figure 6-12 Total number of poultry traded each week by distance and the median number of poultry carried on each journey

†Distance=0 indicates trading within a district

Using the data collected on poultry trading, a conceptual framework of the poultry market chain in Cambodia was developed and is illustrated in Figure 6-13. Poultry is predominantly transported throughout and into Cambodia by middlemen on motorbikes and trucks (Figure 6-14) and are generally traded from more rural sources to more urban/provincial locations (e.g., markets restaurants). Localized poultry movement, i.e., within a rural district, is conducted by middlemen travelling to up to 20 villages each day within a district to purchase poultry. Approximately half of the middlemen interviewed (46.4%) visited at least one village each day with median 3 villages visited per day (IQR: 2-5, Table 6-1) and sold this poultry to markets, largely in Phnom Penh. The remaining middlemen purchased poultry from markets and sold to other markets.

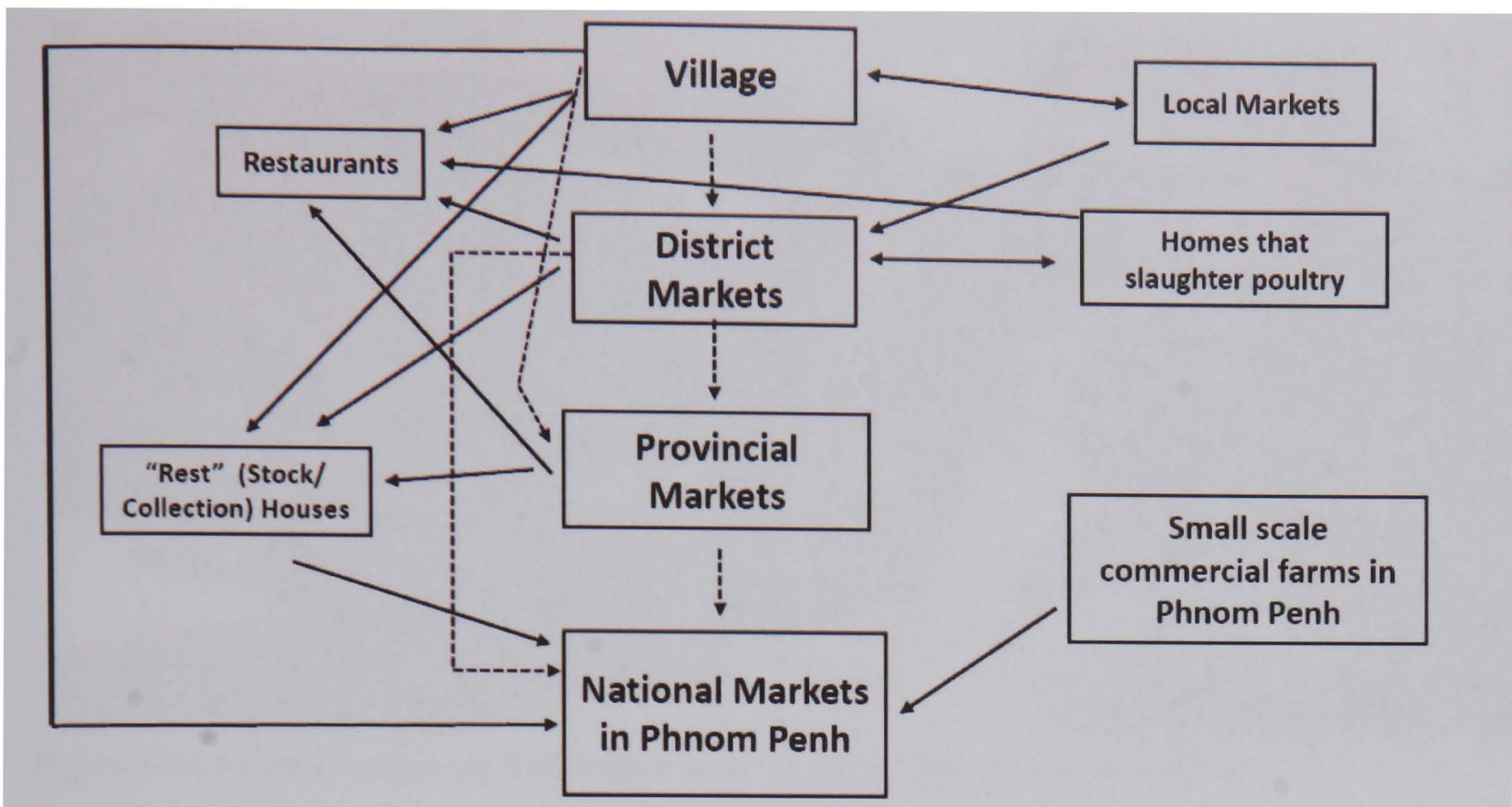


Figure 6-13 Poultry market chain in Cambodia

The arrows represent the movement of poultry via trade. The dotted lines in the figure represent a one-time movement of poultry from one location to a second location. Birds will rarely be traded from their point of origin beyond a second location before slaughter.

Poultry rarely spend more than one day in the market chain before slaughter and commonly move from their point of origin (village or rural market) to a second location (e.g., a provincial/national market) but are rarely traded beyond a second location. For example, it would be unusual for a bird to be traded from a village to a district market and then from that district market via middlemen to national market. Rather birds are sold to markets and on the same day are either purchased by local people for consumption or by local restaurants for slaughter.

Eighty four percent of middlemen reported that they always sold all of their poultry each day. Among market sellers who do not sell all of their poultry during the day 2.1% brought poultry home overnight and back to the market the following day to sell, 54.3% brought them home (to slaughter), 20.2% slaughter them, and 23.4% sold them to restaurants.

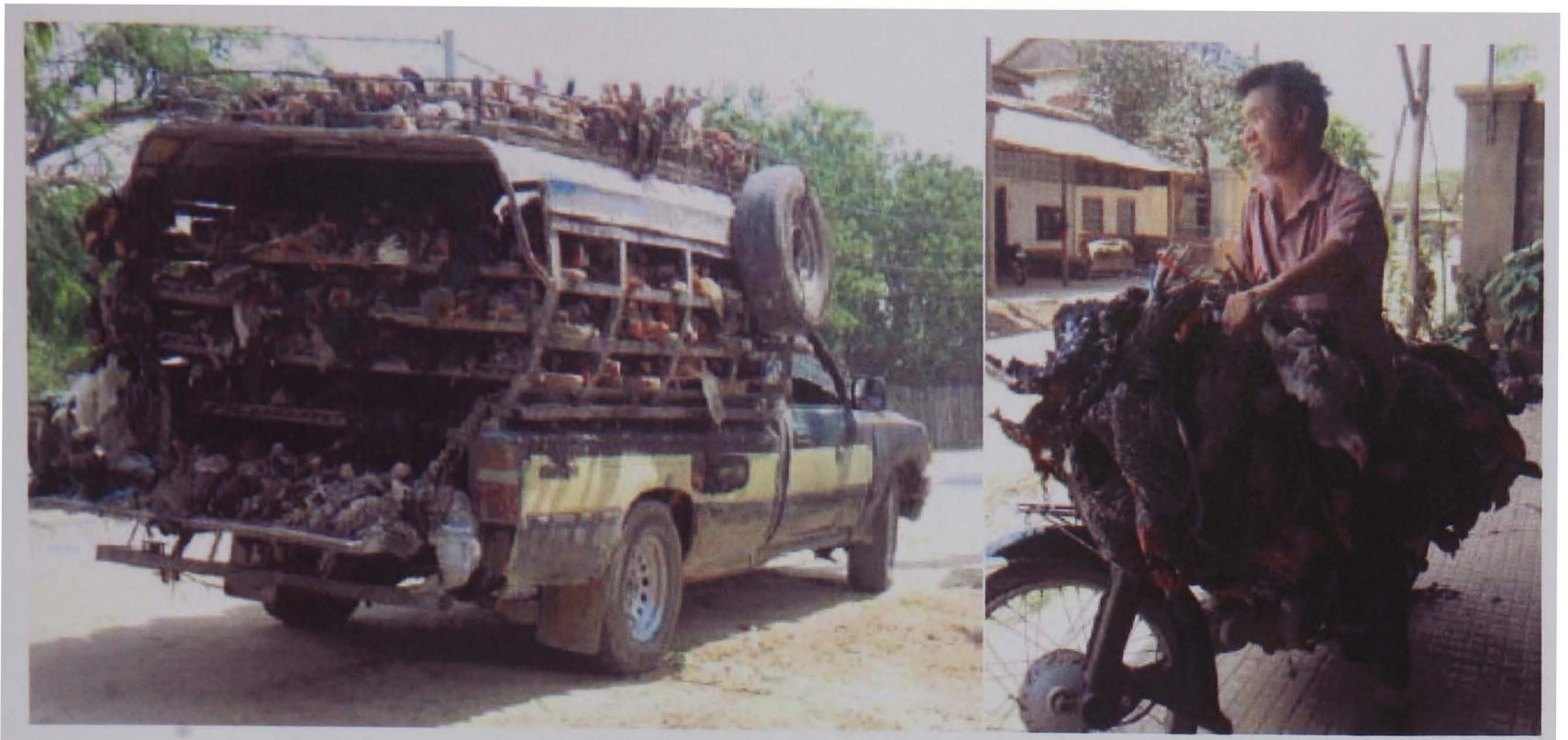


Figure 6-14 Poultry carried via middlemen on trucks (left) and motorbikes (right)

Permission was obtained from all subjects to take and use photographs in reports.

6.3.2.3 *Chicken movement*

With empirical data on weekly poultry selling and transport patterns, directed networks of chicken and duck movements within and into Cambodia were constructed.

Chicken movement is shown in Figure 6-15 and is composed of 133 unique nodes. The network of chicken movement shows great connectivity with 94% of nodes ($n=125$) within a single component. Three smaller isolated components (2-4 nodes each) were not linked with the main network that links to markets in Phnom Penh.

The directed links in the figures illustrate weekly trading of poultry between locations and the median number of chickens traded between any two locations by middlemen each week is 281.3 (IQR: 140.7-410; $n=113$).

The core of the chicken network includes markets in Phnom Penh as well as rest houses and semi-commercial farms in Phnom Penh. The total number of chickens transported throughout the network during an average week of trading (i.e., not prior to a national holiday) is 82,655.

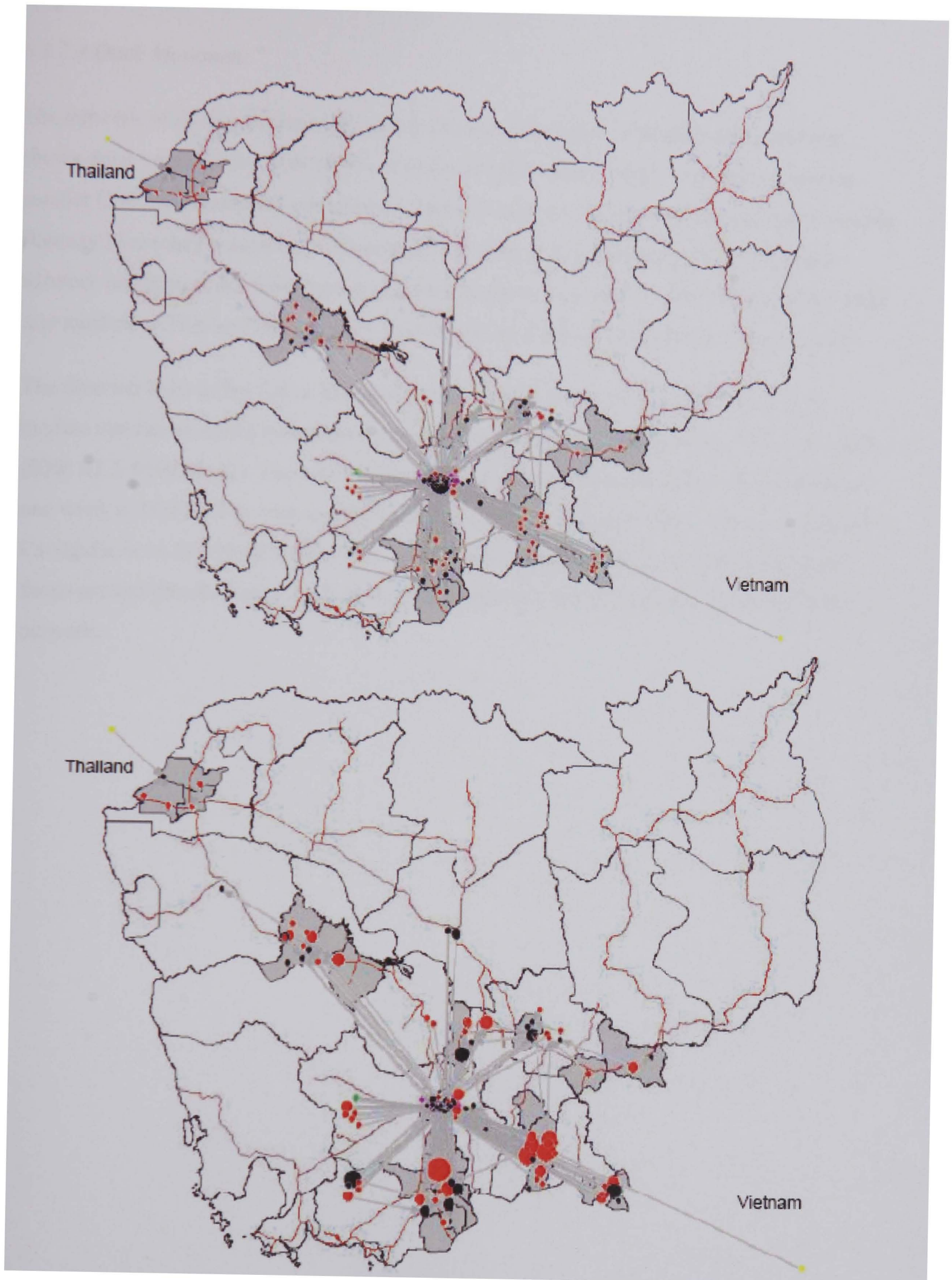


Figure 6-15 Chicken trading network in Cambodia

The figure illustrates (above) node sizes weighted by IN- degree and below the same network weights nodes by OUT-degree. Node colour indicates location type (black=market, purple=stock house, red = rural farm or household, light green = commercial farm, grey=semi-commercial farm, yellow= foreign source), ties show direction as indicated by the arrow and tie strength is indicated by the thickness of the arrow (the thicker the arrow, the more poultry passing between from the two points).

6.3.2.4 Duck Movement

The network of duck movement (Figure 6-16) is composed of 76 unique nodes and also shows great connectivity with 80.3% of nodes (n=61) within a single component, and six smaller (2-4 nodes) isolated components. Isolated networks of duck movement were found in Banteay Meanchey connected to Thailand, in Pursat, and in Kampong Cham. The main network includes nodes from markets from eight provinces and Vietnam and had direct links into markets in Phnom Penh. Poultry from Thailand does not link with the main network.

The directed links in the figure illustrate weekly trading between two locations and the median number of ducks traded between any two locations by middlemen each week is 145 (IQR: 33.3-1000; n=42). The total number of ducks transported throughout the network in one week is 35,049. The total weekly number of ducks transported from sources outside of Cambodia is 16,245 (99.8% from Vietnam) mostly as day old ducks to semi-commercial farms around Phnom Penh, which is approximately half (46.2%) of the ducks in the total network.

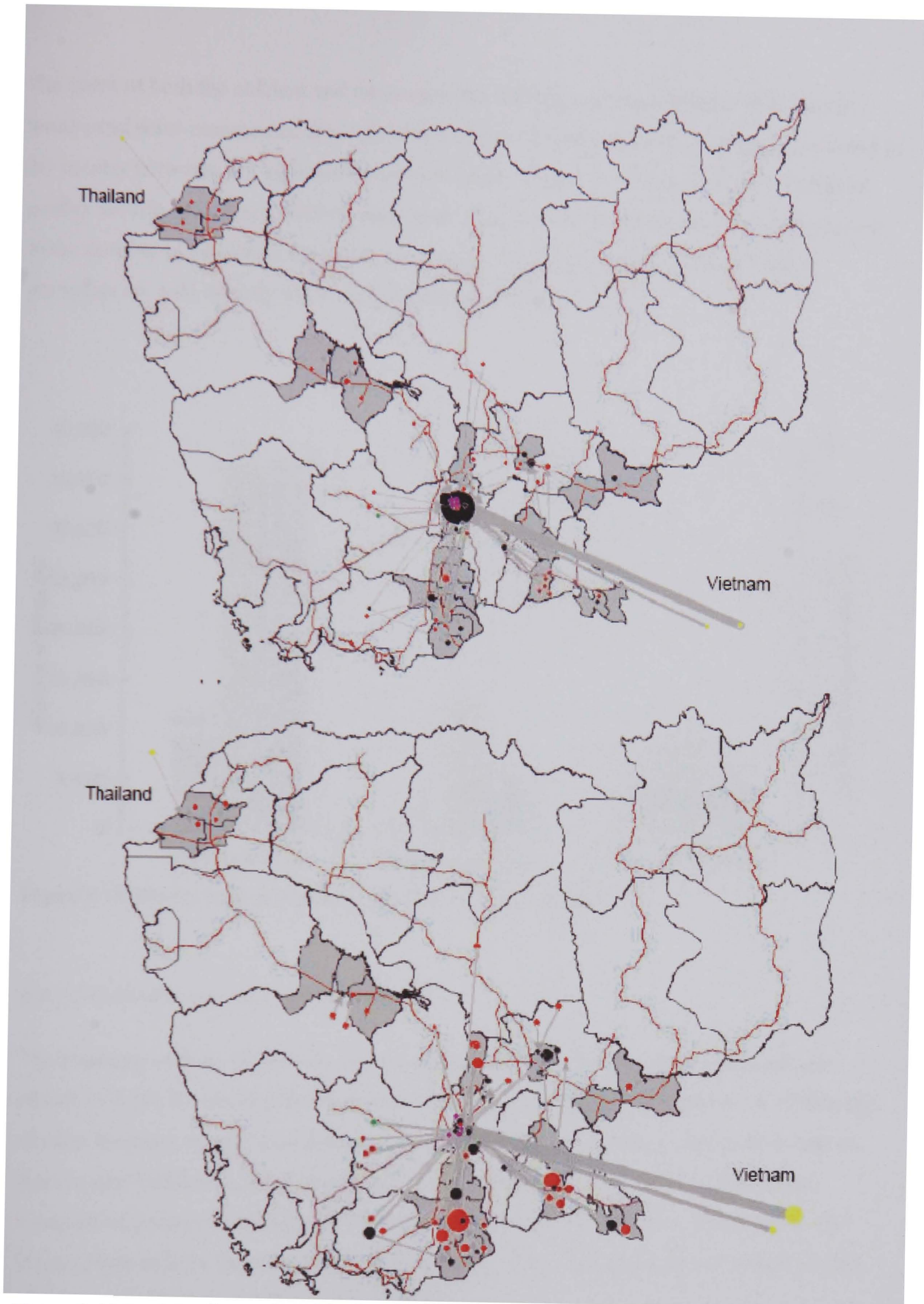


Figure 6-16 Duck trading network in Cambodia

The figure illustrates (above) node sizes weighted by IN- degree and below the same network weights nodes by OUT-degree. Node colour indicates location type (black=market, purple=stock house, red = rural farm or household, light green = commercial farm, grey=semi-commercial farm, yellow= foreign source), ties show direction as indicated by the arrow and tie strength is indicated by the thickness of the arrow (the thicker the arrow, the more poultry passing between from the two points).

The cores of both the chicken and duck networks include markets in Phnom Penh, stock houses and semi-commercial farms in Phnom Penh. The markets with the biggest influence in the poultry networks are all located in Phnom Penh. Figure 6-17 illustrates the quantity of poultry sold to these three markets each week. The secondary axis reports the poultry source as the number of provinces the poultry are traded from. For example, poultry from 11 provinces are sold directly to Orussey market each week.

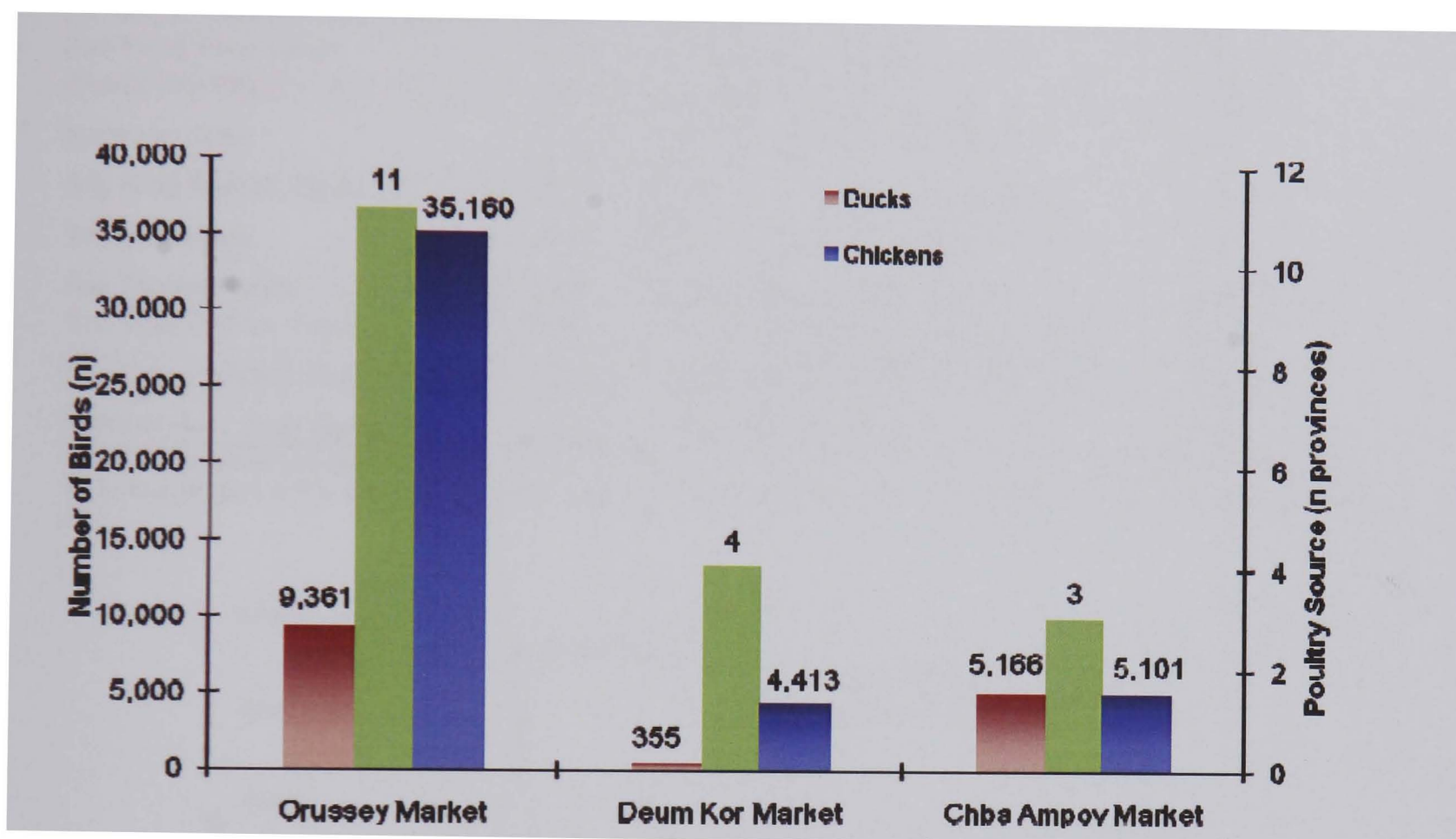


Figure 6-17 Weekly trading of poultry into Phnom Penh markets

6.3.2.5 Centrality measures of the networks

The locations with the highest in- and out-degree measures for the chicken network are shown in Table 6-3 and the distribution of in/out degree is shown in Figure 6-18. Within the chicken trading network, the nodes with the highest out-degree values, that is the locations that are most influential, were from Charoen Pokphand (“CP Company” a Thai based commercial poultry company) and five districts in Prey Veng Province. Three markets in Phnom Penh receive the most chickens (in-degree) each week and include Orussey Market, Chba Ampov Market, and Deum Kor Market.

Table 6-3 Locations with the highest in- and out-degree measures in the chicken network

Location*	Out-Degree	Location*	In-Degree
Chicken Network			
Ang Snuol (CP Company)	9283	Orussey market, Phnom Penh	34742
Ba Phnom District, Prey Veng	6235	Phnom Penh [†]	12519
Prey Veng District, Prey Veng	4777	Chba Ampov Market, Phnom Penh	5101
MeSang District, Prey Veng	3531	Deum Kor Market, Phnom Penh	4413
Takmao District, Kandal	3500	Stock house hear Chba Ampov Market, Phnom Penh	3561
Kampong Trabeak, Prey Veng	3383	Kandal	3500
Pea Rang, Prey Veng	3349	Kandal Market, Kandal	2186
Kandal Province	3347	Kro Kor Market, Pursat	1300
Kampong Speu	3189	Russey Keo (Mean Chay) District, Phnom Penh	1803
Ang Roka Market, Takeo	2388	A Kak Market, Banteay Meanchey	1140
Takeo Province	2229	Stock House near Orussey market, Phnom Penh	844
Ang Tasom, Takeo	2222	Olympic Market, Phnom Penh	801
Tram Kak District, Takeo	2093	Tuol Tompoung Market, Phnom Penh	752
Ang Tasom Market, Takeo	2049	Stock House hear Deum Kor Market, Phnom Penh	700
Romeas Hek , Svay Rieng	1738	Pursat Market, Pursat	620

*The top 15 locations are shown

†A destination listed as “Phnom Penh” was collected from middlemen who were too busy to provide specific market names within Phnom Penh.

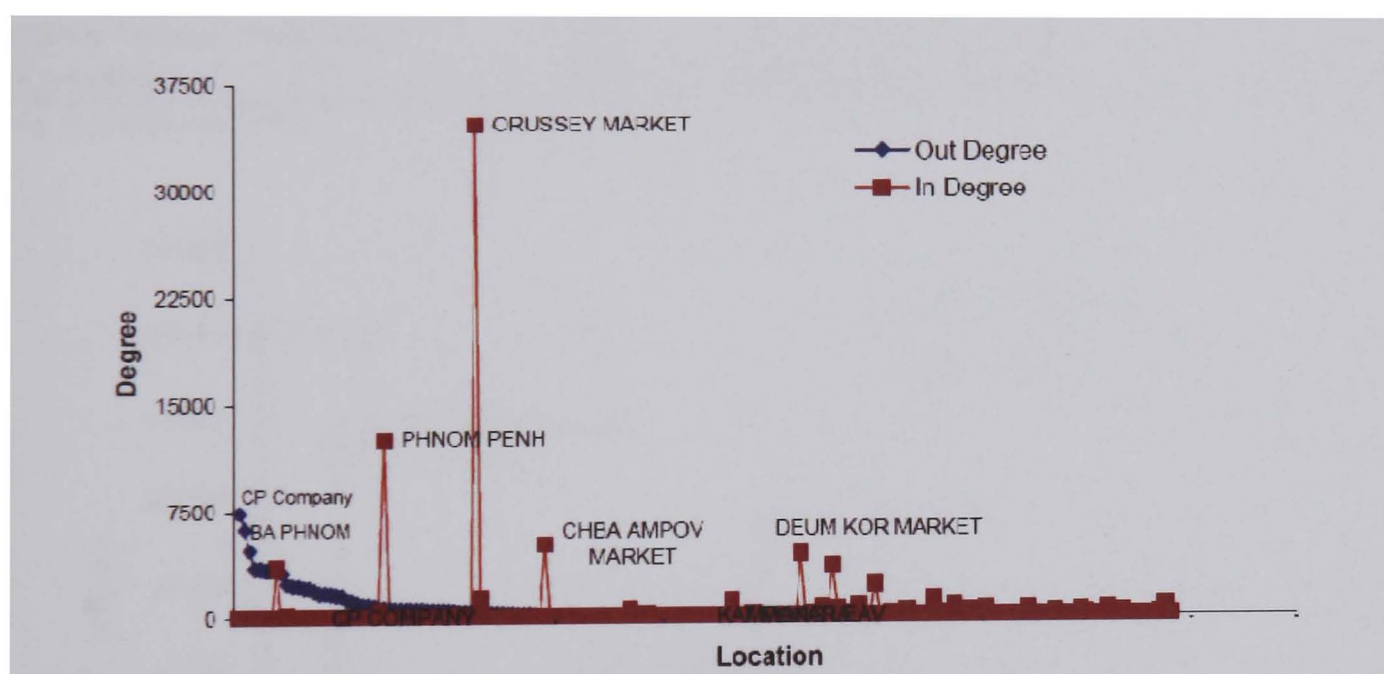


Figure 6-18 In-degree and out-degree of the nodes in the chicken network

Note: The x-axis represents locations and the data provided in the figure has been sorted by out-degree.

The locations with the highest in- and out-degree measures for the duck network are shown in Table 6-4. Within the duck network, the node with the highest out-degree value was located outside of Cambodia (Vietnam); within Cambodia the locations with high duck output include districts in Takeo, Kandal and Kampong Cham provinces.

Locations with the highest in-degree were all located in Phnom Penh and included two main poultry selling markets and semi-commercial farms. The distribution of in/out degree for the duck network is shown in Figure 6-19.

Table 6-4 Locations with the highest in- and out-degree measures in the duck network

Location*	Out Degree	Location*	In Degree
Duck Network			
Vietnam	14110	Russey Keo (Mean Chay) District, Phnom Penh	11400
Tram Kak District, Takeo	2983	Orussey market, Phnom Penh	9317
7 Makara District, Phnom Penh	2500	Chba Ampov Market, Phnom Penh	5166
Prek Phnov Market, Kandal	2400	Dong Kour District, Phnom Penh	3000
Prey Angkor Market, Vietnam	2100	Phnom Penh	1910
Takeo	1709	Takeo	1500
Pea Rang, Prey Veng	1300	Stock house hear Chba Ampov Market, Phnom Penh	813
Batheay, Kampong Cham	1200	Stock House near Orussey market, Phnom Penh	400
Cheung Prey, Kampong Cham	803	Deum Kor Market, Phnom Penh	355
Mean Chey District, Phnom Penh	840	Stock House near 7 Makara market, Phnom Penh	280
Koh Andeok District, Takeo	500	Serei Sophoan, Banteay Meanchey	210
Treang District, Takeo	500	A Kak Market, Banteay Meanchey	145
Prey Veng	488	Ang Tasom Market, Takeo	110
Kampong Trabeak, Prey Veng	365	Ang Roka Market, Takeo	97
CP Company	310	Kro Kor Market, Pursat	60

*The top 15 locations are shown

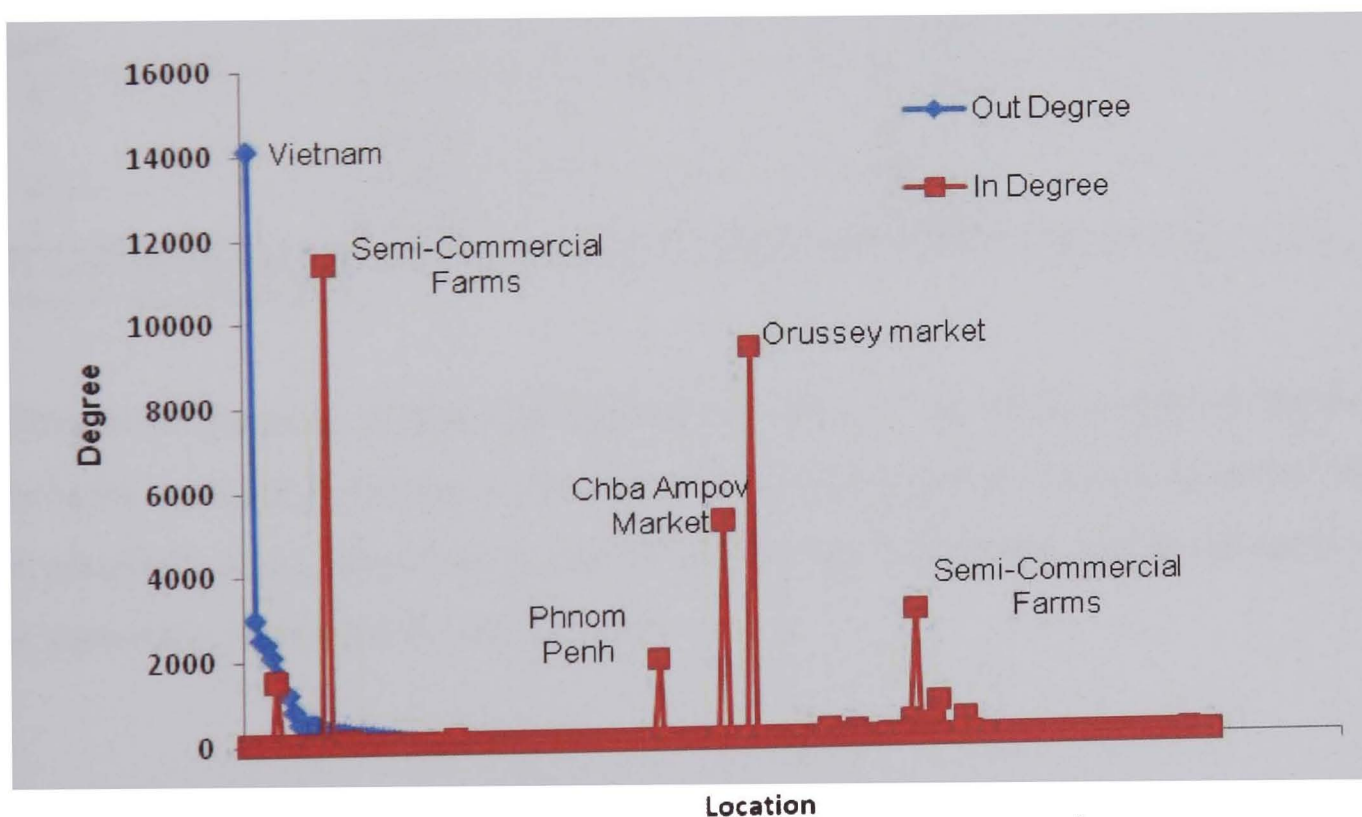


Figure 6-19 In-degree and out-degree of the nodes in the duck network

Note: The x-axis represents locations and the data provided in the figure has been sorted by out-degree.

6.3.2.6 Betweenness of the networks

Because the networks are highly centralized, measures of betweenness are far less important than if the networks contained many intermediary steps before reaching Phnom Penh.

Therefore betweenness were calculated but are not informative in further analyses including in making recommendations for surveillance

6.3.2.7 Core/periphery of the networks

The k-cores of the chicken and duck network are provided in Table 6-5. The k-core indicates those nodes with at least k connections. In the duck network, for example, there are four nodes with four or more connections, seven nodes with three connections, 19 nodes with two connections and the majority of the nodes in the network have one connection (60.5%). In the chicken network, more than half (52.6%) of the nodes have only one connection to another node, whereas 21.1% have two connections, 15% have three connections and 11.3 have four or more connections.

Table 6-5 Number of connections of each node in the networks

K-core	Network	
	Duck (Total Nodes = 76) n (%)	Chicken (Total Nodes = 133) n (%)
4	4 (5.3)	15 (11.3)
3	7 (9.2)	20 (15.0)
2	19 (25)	28 (21.1)
1	46 (60.5)	70 (52.6)

K-core is the number of connections to each node: e.g., k=core = 1, means the node has only 1 connection, 2= 2 connections, 3= 3 connections, 4= at least 4 connections.

For illustrative purposes, Figure 6-20 presents an image of the chicken network highlighting nodes in the core and periphery of the network. Nodes around the outside represent locations in the periphery of the network (i.e., low k-core scores), while nodes inside represent the core of the network (i.e., highest k-core scores).

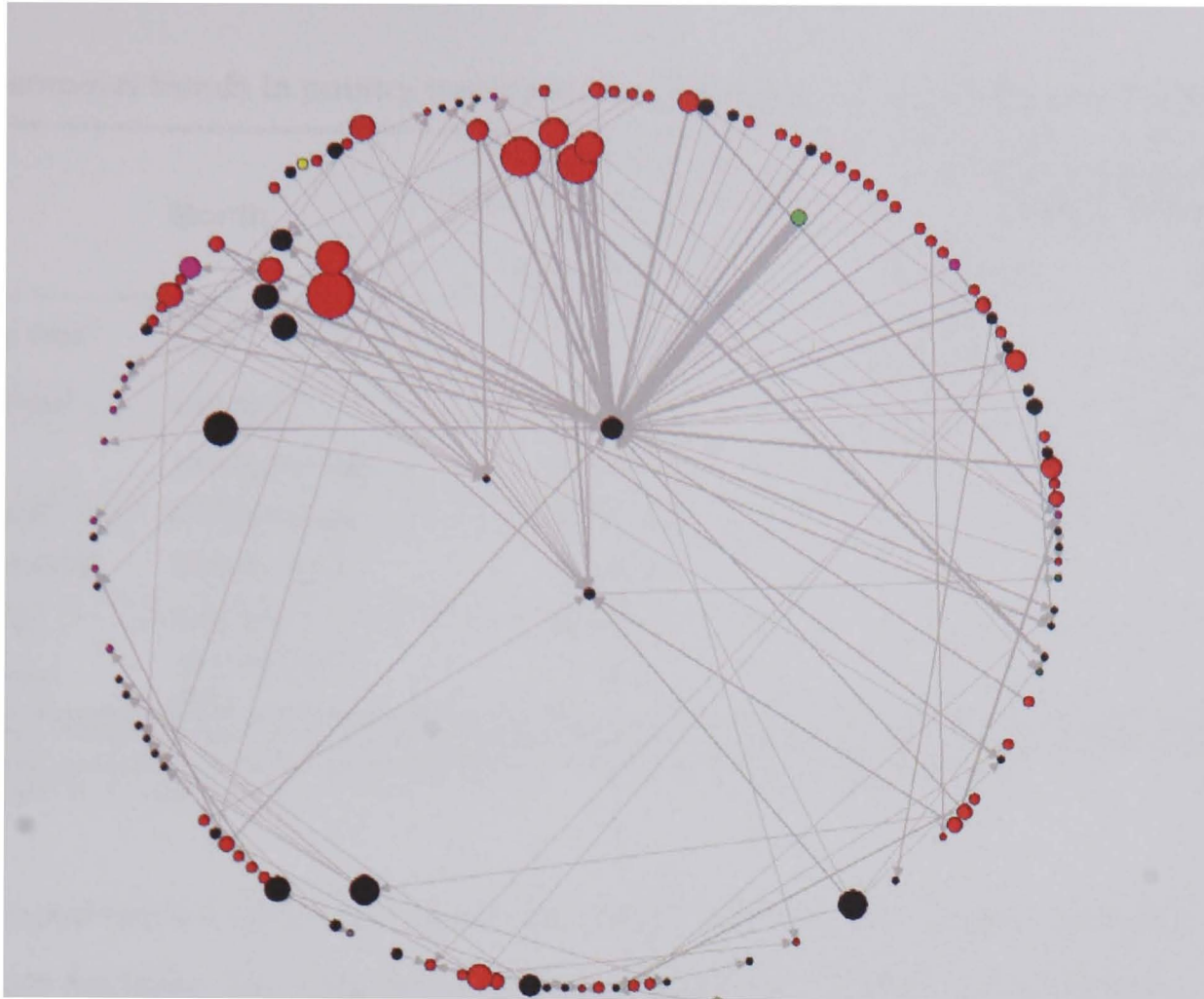


Figure 6-20 Core and periphery of the chicken network

This network is identical to the chicken trading network shown in Figure 6-15 with nodes weighted as out-degree. Nodes in the inside represent nodes within the core of the network and represent markets in Phnom Penh, CP Farms, rural locations in Prey Veng and markets and rural locations in Takeo, while nodes along the edge represent nodes in the periphery of the network.

Node colour indicates location type (black=market, purple=stock house, red = rural farm or household, light green = commercial farm, grey=semi-commercial farm, yellow= foreign source), ties show direction as indicated by the arrow and tie strength is indicated by the thickness of the arrow (the thicker the arrow, the more poultry passing between from the two points).

6.3.3 Seasonality of Poultry Movement

Several annual festivals were noted among market sellers and middlemen as increased periods for poultry trade. The two main annual holidays are Chinese New Year, which occurs each year in late Jan/early Feb and the Khmer New Year, which occurs in each year in mid-April. Subjects also noted several other holidays where poultry trade increases (Table 6-6).

Table 6-6 Temporal trends in poultry trading by market sellers and middlemen in Cambodia

Occasion	Month	Subjects reporting occasion as increased poultry trade n (%) (n=262)	% Median Increase (IQR) in Poultry Trade	
			Chickens	Ducks
Chinese New Year ^a	late Jan/early Feb	204 (77.9)	122.2 (48.5-170.7) [†]	50 (31.2-921.7) [‡]
Khmer New Year ^b	mid-April	71 (27.1)	97.5 (89.9-197.7) ^{††}	1265 (636.5-1894.1) [*]
Pchum Ben ^c	15 days in Sept	36 (13.7)	--	--
Sen Kbal Teuk ^d	1 day in Sept	43 (16.4)	--	--
"Wedding" Season ^e	October-April	46 (17.6)	--	--
Chheng Meng ^f	April 1- 5	40 (15.3)	--	--
Islam New Year ^g	late Dec/early Jan	1 (0.4)	--	--

^a Lunar New Year; ^b Cambodian New Year; ^c Cambodian ceremony offering food to spirits; ^d Chinese ceremony offer food to spirits; ^e Period of increased occurrence of weddings due to cooler weather; ^f Offering to the spirits; ^g Islamic New Year
[†]n=47; [‡]n=6; ^{††}n=7; ^{*}n=2; -- not available

Middlemen and market sellers reported higher periods of trade during the 1-4 weeks prior to these festivals. The links and direction of poultry movement do not change during these festivals (i.e., the networks do not change). However, the volume of birds traded each week increases.

For example, middlemen reported a median increase in chicken and duck trading of 122.2% (IQR: 48.5 - 170.7; n=47) and 50.0% (IQR: 32.3 - 921.7; n=6), respectively, during the weeks prior to the Chinese New Year, and a median increase in chicken and duck trading of 97.5% (IQR: 89.9-197.7; n=7) and 1,265.8% (IQR: 636.5-1895.1; n=2), respectively, during the weeks prior to the Khmer New Year.

6.3.4 Potential for HPAI Viral Spread Across the Poultry Networks

Both the chicken and duck networks showed a high degree of connectedness and centrality with directional movements from rural areas into nodes in Phnom Penh. Figure 6-21 illustrates the potential spread of H5N1 in four hypothetical outbreaks, each with different locations where the outbreak was seeded (indicated by the black circle). The red nodes indicate the potential spread of the disease via directed trade links between the nodes. Node shape differentiates between premises type (e.g., rural source, market, semi-commercial farm, etc).

Figure 6-22 summarizes results of seeding the outbreak in various nodes throughout the network and the spread via the connectedness of the trade links. The figure shows the total number of locations potentially infected based on where H5N1 is identified.

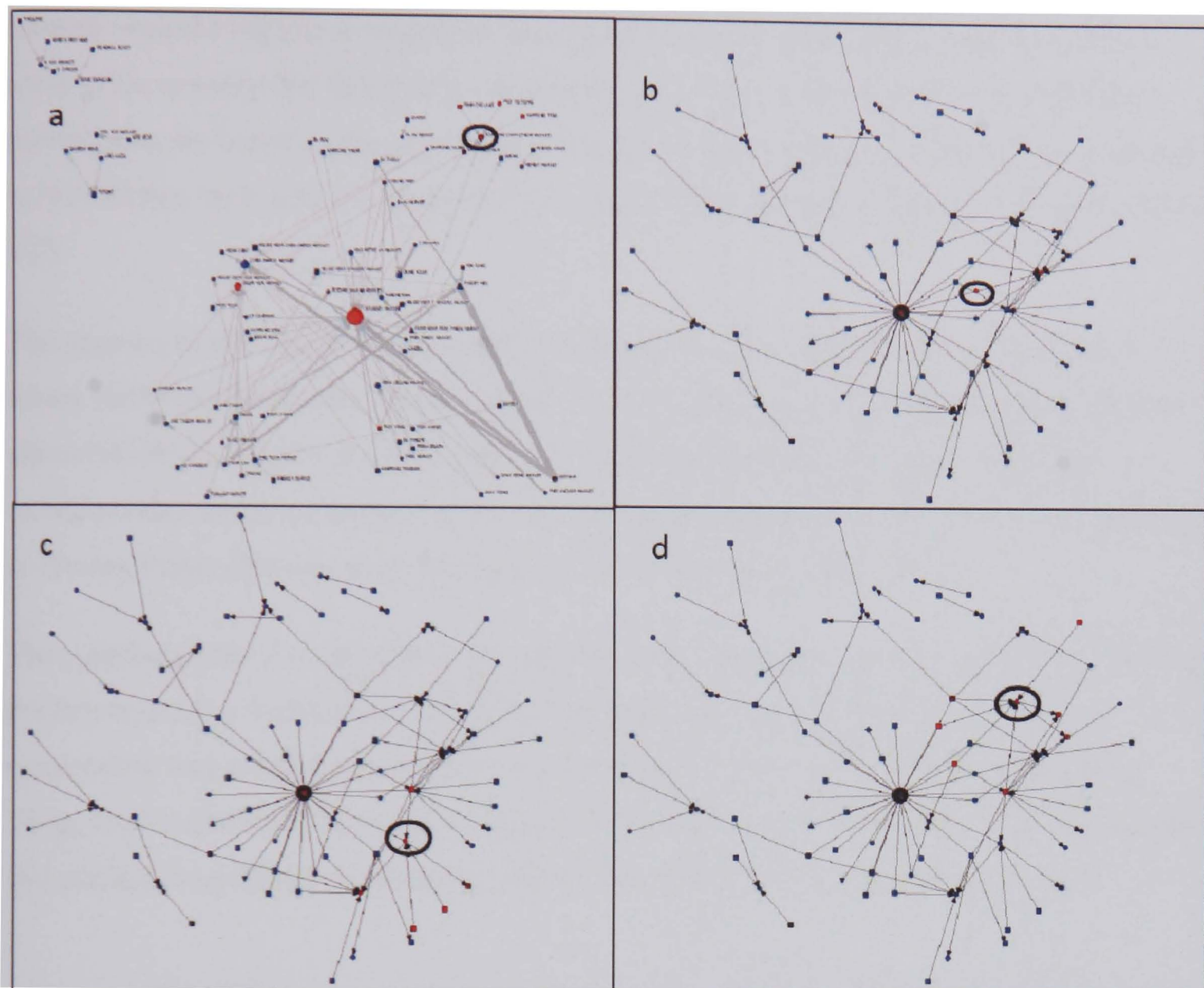


Figure 6-21 Connectedness of the network: a-d) Potential for disease spread across duck network by seeding the outbreak in a) Kampong Cham Market b) Svay Rieng Market c) Ang Roka Market, Takeo Province and d) a semi-commercial farm in Phnom Penh

The red nodes indicate infected premises; blue node uninfected; node shape differentiates between premises type (e.g., circle =market, square = rural source, triangle= semi-commercial farm)

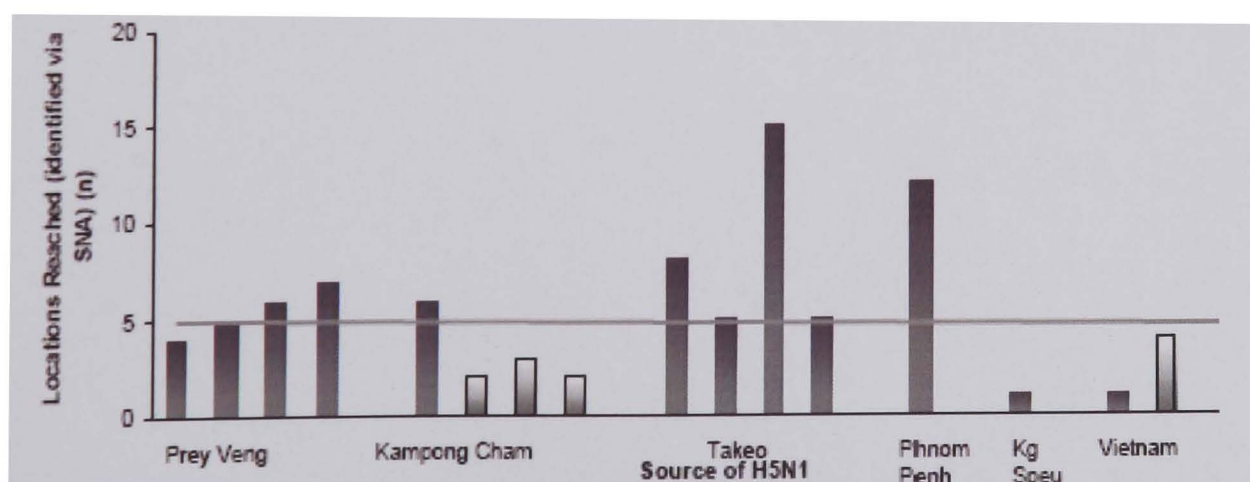


Figure 6-22 The number of locations potentially infected based on the location of H5N1 identification

The horizontal line = median value 5

6.3.5 Risk-Based Surveillance Recommendations

Since the structures of the poultry networks in Cambodia are highly centralized and represent a spoke-and-wheel type structure with unidirectional movement into Phnom Penh, I identified markets with the highest in-degree as those most likely to detect HPAI if present in many areas of the country due to the large catchment area. The recommendations for risk-based surveillance are based on the objectives of the surveillance system and I have recommended to NaVRI that their active surveillance activities of HPAI be prioritized into two tiers (Table 6-7).

The purpose of the Tier 1 recommended locations for surveillance is to identify locations where HPAI can be rapidly detected if the virus is in the market system and would indirectly allow NaVRI to monitor the HPAI status of poultry populations in rural areas. These recommended locations include those with the highest in-degree values, and include markets in Phnom Penh, semi-commercial farms and stock houses in Phnom Penh.

The purpose of tier 2 recommendations is to identify locations that are most likely to interrupt poultry-to-poultry transmission of HPAI. These are locations with greatest number of connections and the highest out-degree scores. Using these criteria, four districts in Prey Veng, four markets in Takeo, one market in Kampong Cham, one market in Kampot province and poultry from CP farms should be included in HPAI active surveillance activities.

Table 6-7 Recommended priorities for active HPAI surveillance in Cambodia

Location	Description	Justification for inclusion in surveillance
Tier 1 Purpose: To identify locations where HPAI can be rapidly detected if in the market system		
1 Markets in Phnom Penh: Orussey Market, Chba Ampov Market and Deum Kor Markets	Markets with the greatest numbers of poultry trading, largest wet markets in Phnom Penh	Highest in-degree, most connections within both the chicken and duck networks
2 Semi-commercial Farms in Russey Keo District, Phnom Penh	16 semi-commercial farms near Phnom Penh, which have holdings of 2,000-4,500 chicken or 400-2,000 ducks; Ducks are day old ducks from Vietnam and unvaccinated for H5N1	
3 Stock houses near major poultry markets in Phnom Penh	Approximately 12 stock houses that serve as an intermediary between middlemen from provinces outside of Phnom Penh and the markets in Phnom Penh. [†] Owners keep live poultry in cages for less than 8 hrs before selling onto markets. Some stock houses slaughter poultry before bringing to markets.	
Tier 2 Purpose: To identify locations that are most likely to interrupt poultry-to-poultry transmission		
1 Prey Veng: Prey Veng, Ba Phnom, MeSang, Kampong Trabeak	Most chickens come from rural sources in these 4 districts Prey Veng, most ducks come from Prey Veng Market	Nodes with greatest number of connections and the highest out-degree scores.
2 Markets in Takeo: Takeo Market, Tram Kak Market, Ang Roka Market, Ang Tasom Market	Most ducks come from these markets in Takeo and are traded to markets in Phnom Penh	
3 Market in Kampong Cham: Kampong Cham (Thom) Market	Most ducks come from this markets in Kampong Cham and are traded to markets in Phnom Penh	
4 Market in Kampot: Chhouk Market	Most chickens and ducks come from this market in Chhouk into Phnom Penh.	
5 CP Company	Commercial poultry facility with locations in Kampong Speu and Kandal Province‡	

[†] Further research is needed to identify ALL stock houses in Phnom Penh

[‡] Further research is needed on CP farms

6.4 Discussion

My results have demonstrated that live poultry movement in Southern Cambodia is unidirectional, highly connected and centralized. I found that the premises involved in poultry trade are closely linked via middlemen carrying live poultry over long distances and that the unidirectional movement of poultry into Phnom Penh make these markets in Phnom Penh a potential hub for the spread of H5N1, and thus ideal for HPAI surveillance and control.

Approximately half of the live ducks and most live chickens entering into Phnom Penh come from rural areas of Cambodia with backyard poultry holdings (Sector 4 poultry production (FAO 2006)). Localized movement, i.e., <5-10 km radius of the source, was not fully captured by the network analysis as rural Cambodians are not in the regular habit of selling poultry outside of their villages. Rather backyard poultry are entering the market chain by middlemen who reported visiting up to 20 villages a day to obtain enough poultry to sell to a market. This pattern of multiple and routine visits to villages by middlemen presents a major limitation of the control and surveillance of HPAI in Cambodia because their vehicles could serve as the potential mechanism to spread the virus from village to village before they reach markets or from markets back to their home village if they visited an infected market.

Approximately 85% of middlemen trade live birds >10 km from where they purchased the animals. Guidelines in the European Union and United States recommend restricting the movement poultry for the control of H5N1 (2006; USDA/APHIS), a policy that would likely be difficult to administer in Cambodia and unlikely to fully stop poultry movement into Phnom Penh.

Poultry movements within the province bordering Thailand (Banteay Meanchey) and into this province from Thailand are separated from the main network linking Phnom Penh. Of importance to highlight, however, is the dominance of ducks traded from Vietnam into the Cambodian market chain. Cross-border movement of poultry is currently illegal, however this research identified more than 16,000 ducklings entering the Cambodian market chain weekly, composing the other half of the total duck network. Ducks from Vietnam are sold directly to semi-commercial farms and markets located in Phnom Penh, all of which should be included in routine HPAI surveillance activities.

Though this study was not able to capture localized cross-border trading activities by rural Cambodians, it is possible that low levels of trading poultry across international borders is occurring (IPC Unpublished Data) and likely at an increased rate around the time of the Chinese and Khmer New Year festivals when there may be a large differential between the cost of poultry on either side of the border. Although the extent of this trading is unknown, I am aware of several incidents of illegal trading of live chickens and ducks across the border around the Chinese and Khmer New Years (MAFF Unpublished Data).

This is further supported by the remarkable increase that is reported in live poultry trading via market sellers and middlemen in the weeks prior to the Chinese and Khmer New Years festivals. Although my results are based on small sample sizes, the magnitude of the increase indicates that there are significant increases in trade volume during the weeks prior to these holidays. All of the human cases and approximately half of the domestic poultry outbreaks in Cambodia have occurred between the Chinese and Khmer New Year festivals (OIE 2008a; WHO 2006-2009), times in which the consumption of poultry increases. Increases in H5N1 poultry outbreaks in Nigeria in 2006-2007 may have been linked to increased periods of trading (Joannis et al. 2008).

My results have also identified the need to improve the working conditions of poultry selling markets, which can be greatly enhanced by implementing basic improvements in the hygiene of the markets and the use of protective equipment by market sellers and by individuals responsible for slaughtering poultry at the markets. Although I found that poultry spend *at most* one day in the market chain before slaughter, the conditions in the live birds selling areas and slaughtering areas could facilitate a viable environment for HPAI to survive and/or persist (Kung et al. 2007; Kung et al. 2003b; Nguyen et al. 2005; Webster 2004). Since the implementation of rest days in live bird selling markets has been successful in markets in Hong Kong (Kung et al. 2003b) and recent evidence from NaVRI has identified asymptomatic ducks with H5N1 antibodies in several markets in Cambodia (MAFF Unpublished Data), implementing routine monthly disinfection in the wet markets with the largest influx of poultry might be important in reducing the potential for animal-to-animal and animal-to-human HPAI transmission.

As of 25 July 2008, 1,137 H5N1 outbreaks in domestic poultry have been confirmed in Thailand, 2,490 in Vietnam and 22 in Cambodia (OIE 2008e). There are several possible explanations why H5N1 is not more widely spread within Cambodia. First, the estimates of poultry density in Cambodia and neighbouring countries vary significantly. Poultry density is believed to be highest in Thailand (approximately 509-1,637 poultry per km²) followed by Vietnam (approximately 663-787 poultry per km²), Cambodia (approximately 99-175 poultry per km²) and Laos PDR (approximately 73 poultry per km²) (Burgos 2008; Dung 2007). It may be possible that poultry density in Cambodia is not high enough to sustain virus circulation. However it is more likely that the structure of Cambodia's poultry production system with predominant free-ranging backyard poultry ownership and lack of semi-commercial farms throughout the country are limiting the circulation of HPAI (Chapter 4). In addition it is likely that asymptomatic HPAI infection in duck flocks may have occurred in addition to underreporting of poultry mortality due to HPAI in rural regions of Cambodia (Chapter 4).

Secondly, at present, it is difficult to determine the probability that a diseased bird will enter into the market chain in Cambodia. Nearly all (99%) middlemen I interviewed stated that they do not trade visibly sick poultry, which does not exclude introduction of asymptomatic but infectious ducks in the market chain. Furthermore, rural households experiencing poultry mortality due to illness typically prepare sick or dead poultry for household consumption or give away to neighbours within their village rather than selling to a market or middleman (Chapter 4) making it possible that an outbreak would occur within or between villages with limited spread.

As with all cross-sectional surveys, it is possible that I did not capture all relevant poultry movements. The high connectivity I found in the poultry networks could be the result of my sampling frame as I did not survey all markets within Cambodia, particularly in Northern Cambodia. I do believe, however, that this study has captured a fairly complete network of live poultry movement in study areas and their connections into and out of Phnom Penh.

Furthermore, I am confident that this study has identified the bulk of live poultry movement into Phnom Penh because, in addition to sampling markets and 115 villages in 24 districts in six provinces, I interviewed middlemen on multiple occasions along all six national roads into Phnom Penh, at stock houses located next to the three main poultry selling markets, at semi-

commercial farms in Phnom Penh and at markets in Phnom Penh. This study did not find poultry trade links with Siem Reap (Northwest Cambodia) or areas in the far-western and north-eastern provinces (areas that are highly mountainous and sparsely populated) of Cambodia. Thus while my network does not represent movement in these areas, I am confident that these poultry do not reach Phnom Penh.

Given the rapid global spread of HPAI/H5N1 in recent years, surveillance of poultry populations will remain a high priority, particularly in the Mekong Delta Region where a considerable number of human deaths have occurred. This study has been able to identify critical points for active HPAI surveillance and has informed Cambodia's HPAI surveillance activities. However this does not replace the need for passive surveillance, which should be strengthened in rural areas of Cambodia by encouraging poultry owners to report any and all poultry mortality to village animal health workers and their village chiefs.

Since active surveillance in markets is likely to remain a component of the surveillance and control efforts for HPAI in Cambodia and elsewhere, my results can be used to inform the selection of markets that best suits particular objectives of the surveillance system, in particular whether the objective is monitoring of the HPAI status of poultry populations in rural areas or early detection of incursion in markets with high potential for spread. Collection of similar data in other countries could prevent outbreaks or incursions of HPAI within their borders.

Chapter 7 Fitting Gravity Models to Poultry Movement Data in Cambodia

7.1 Introduction

Previous studies have demonstrated that the connectedness of animal networks can result in large and widespread epidemics of disease (Dent et al. 2008; Green et al. 2008; Kiss et al. 2008; Truscott et al. 2007).. In Chapter 6, I showed how knowledge of poultry movement networks can be used to design more targeted surveillance activities and inform models of disease spread. My results demonstrated that poultry movement in the research study areas is highly connected, unidirectional and centralized. In this Chapter a gravity model is fit to these poultry movement data using population data as an indicator of potential trade between the source where poultry are reared and destination of where poultry are sold to attempt to understand the potential driving forces behind the poultry movement patterns observed.

Gravity models have been used to describe and predict movement based on the characteristics of, for example, economic size as defined by GDP, income level or population size of two locations and the distance between two locations, and have been largely utilized in transportation planning (Erlander & Stewart 1990) and in the field of economics to predict international trade flow (O'Kelly 1999). The general theory of gravity models is represented as

$$C_{ij} = \theta \frac{N_i^\beta N_j^\epsilon}{d_{ij}^\gamma}$$

where C_{ij} represents workflow or trade between i and j , d_{ij} is the distance between location i and j ; N is a characteristic of location i or j (e.g., population size), θ is a constant and the exponents β , ϵ and γ are model parameters (Xia et al. 2004).

Few examples of the use of gravity model theory outside of economics and transportation planning exist. However, gravity models have been applied infectious diseases to evaluate the relationships between disease spread, population sizes and distance (Viboud et al. 2006; Xia et al. 2004). For example, Viboud et al. 2006 use a gravity model to describe transmission of

influenza between states by fitting such a model to work flow data, i.e., movement and distance between home and work (Viboud et al. 2006). In their model they assume that movement predicted by the model is proportional to county population size and Euclidean distance between the residence and work county centres:

$$Flow_{Residence \rightarrow work} \propto \frac{P_{Residence}^{\beta} P_{work}^{\epsilon}}{d_{residence,work}^{\rho}}$$

Using predicted workflow between states as calculated by the gravity model, the authors simulated the spread of influenza across states and compared the epidemic spread predicted by the gravity model to real epidemics of seasonal influenza. They found that epidemic spread is more rapid between well connected states (e.g., California) as compared to more isolated and less populated states (Viboud et al. 2006). Similarly, Xia et al. used a gravity model motivated by their use in modelling population movement and then fit the parameters of the model to match the spatio-temporal dynamics of measles outbreak data in the UK (Xia et al. 2004). They combine a gravity model:

$$Movement(j, k) = \frac{N_{k,t}^{\beta} N_{j,t}^{\epsilon}}{d_{jk}^{\rho}}$$

where d_{jk}^{ρ} is the distance between communities (distance measure not provided) and N_k and N_j are the population sizes of community k and j at time t , with a transmission model of measles using weekly case reports for approximately 1000 communities in England and Wales, to predict the spatial characteristics of measles (Xia et al. 2004).

Both studies found that gravity model theory was useful in predicting epidemic spread based on distance between communities (or states) but found that other factors underlying host movement (e.g., age of those moving between communities) were important to understand local as compared to long range spread (Viboud et al. 2006; Xia et al. 2004). Viboud et al found that the exponent parameter estimates for population sizes for both the residence and work countries were less than 1 indicating that individuals in small populations (e.g., cases of seasonal influenza) may be proportionately more important for disease spread.

The main advantage of using a gravity model within transmission models rather than relying on the underlying movement data is that it can be applied outside the study areas. Thus, by fitting a model to the poultry movement data from Cambodia, it may be possible to predict trade flows in areas not covered by the study as well as in the wider Mekong Delta Region which would be informative for HPAI control programs. Furthermore, as gravity models use information on the underlying populations, they should in theory be able to predict changes in movement patterns following underlying changes in the population, although such predictions have yet to be validated within an infectious disease context.

7.1.1 Objectives of the Chapter

The objective of this chapter is to

- Develop a series of spatial models that describe the driving forces behind poultry movement in Cambodia; and
- Evaluate the fit of the spatial models to the poultry movement data collected in this study.

7.2 Methods

7.2.1 Model Structure

The model assumes that the movement of poultry is determined by a function of the distance between the two locations (the spatial kernel) and the populations at the source i and destination j . Thus the flow of poultry at distance d_{ij} , $f(d_{ij})$ is given by the equation:

$$f(d_{ij}) = GN_i^\varepsilon N_j^\beta k^*(d_{ij})$$

where N_i is an attribute of source location i (here either the human or poultry population), N_j is an attribute of destination location j , $k^*(d_{ij})$ is the normalised spatial kernel, ε and β are parameters which scale the influence of the source and destination populations, respectively, and G is a scaling parameter.

I considered two potential functions for the spatial kernel. The first was that the flow of poultry would decline exponentially with distance and thus the spatial kernel $k(d_{ij})$ is given by:

$$k(d_{ij}) = \exp(-d_{ij} / \lambda)$$

where λ is the kernel parameter. This is illustrated in Figure 7-1

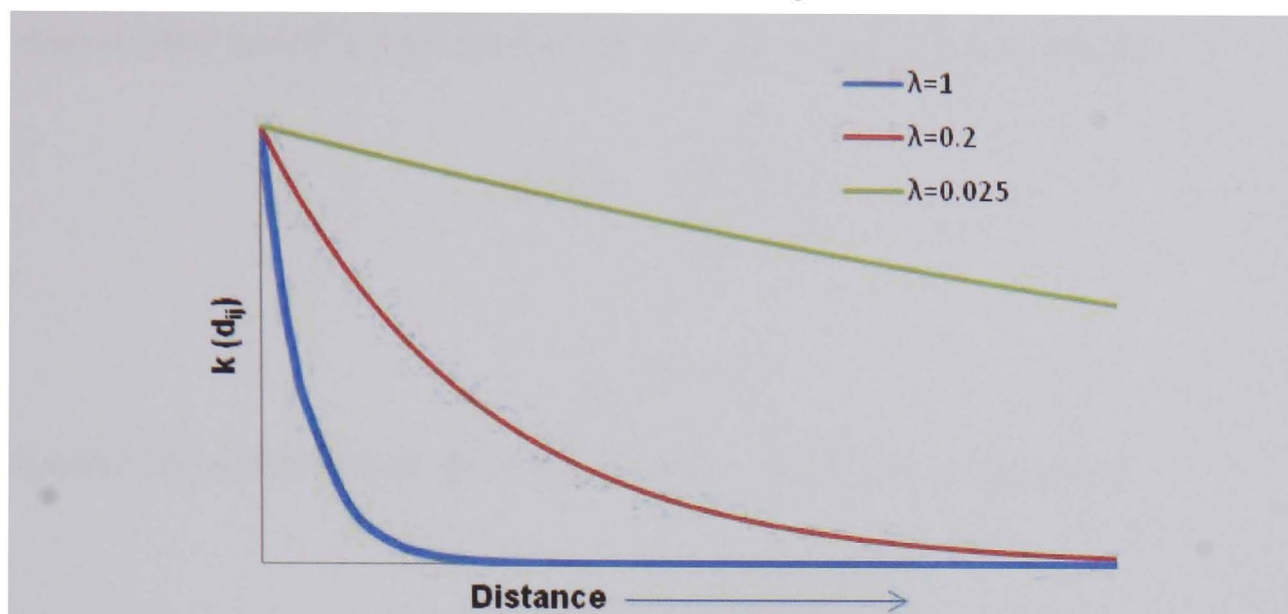


Figure 7-1 The probability of movement declines exponentially with distance

(parameters $\lambda=1, 0.2$ and 0.025 and Distance $0-20\text{km}$)

The second was a power law function which has been used extensively in other disease models (e.g., (Chis Ster & Ferguson 2007; Ferguson et al. 2005)) with the spatial kernel given by:

$$k(d_{ij}) = \left(1 + \frac{d_{ij}}{\lambda}\right)^{-\gamma}$$

where λ and γ are kernel parameters. This is illustrated in Figure 7-2.

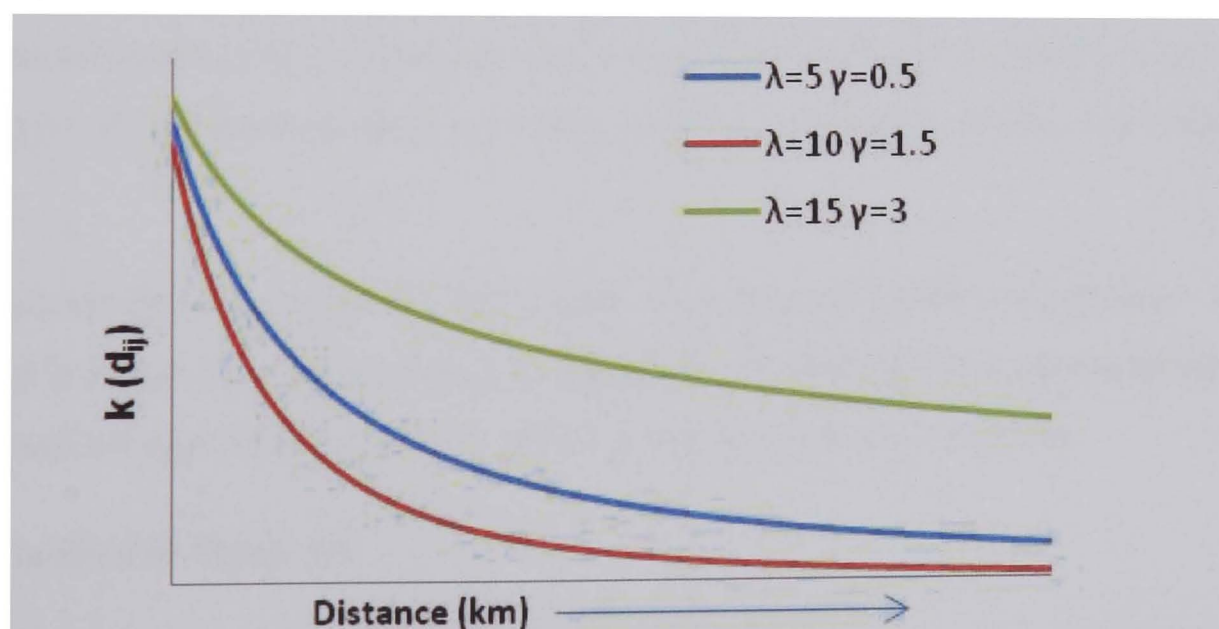


Figure 7-2 The probability of movement as a function of distance as given by a power law function

For parameters $\lambda=5, 10, 15$ and $\gamma = 0.5, 1.5, 3.0$ and distance $0-50 \text{ km}$

The normalised kernel $k^*(d_{ij})$ was obtained by dividing the spatial kernel by its integral over two-dimensional space up to a maximum distance D (assumed here to be equal to 200,000 km)

For the exponential spatial kernel the bottom term of this equation is given by:

$$k^*(d_{ij}) = \frac{k(d_{ij})}{\int_0^{\infty} r e^{-\lambda r} dr}$$

where:

$$\int_0^{\infty} r e^{-\lambda r} dr = \lambda^{-2}$$

For the power law spatial kernel the bottom term of this equation is given by:

$$k^*(d_{ij}) = \frac{k(d_{ij})}{\int_0^D r k(r) dr}$$

where:

$$\int_0^D r k(r) dr = \frac{\lambda}{(1-\gamma)(2-\gamma)} \left[\lambda - \left(1 + \frac{D}{\lambda}\right)^{(1-\gamma)} (\lambda + (\gamma-1)D) \right]$$

As poultry population data were not initially available, I first considered using human population data as an indicator of potential trade between the source and destination. Thus under this model I let $N_i=H_i$ and $N_j=H_j$ where H_i and H_j are the human population estimates at locations i and j respectively. After obtaining poultry data from ministry officials at NaVRI, I considered an alternative model where the supply from location i would depend on the number of poultry at $N_i=P_i$, while the demand would depend on the number of people at destination $N_j=H_j$.

Thus after poultry data were obtained, the models were fit using estimates of poultry population at the source (supply) and human population at the destination (demand) separately using a normalized exponential function and a normalized power law function.

7.2.2 Evaluation of Model Fit

The fit of the models was evaluated using the deviance statistic:

$$\text{Deviance} = -2 * (\text{LogLikelihood (model)}) - (\text{LogLikelihood (saturated model)})$$

Under a Poisson likelihood this is given by:

$$D \propto -2 \sum_{i,j} \left[O_{ij} \ln(E_{ij}) - E_{ij} - O_{ij} \ln(O_{ij}) + O_{ij} \right]$$

where O_{ij} is the observed poultry (chickens and ducks) traded between locations i and j as collected from the questionnaires of market sellers and middlemen and E_{ij} is the expected poultry traded as calculated by the model, $E_{ij} = f(d_{ij})$. The solver function in Excel was utilized to minimize the deviance by changing the kernel parameter values (γ, λ), power parameters (ϵ, β) and the scaling parameter (G).

The model fit was assessed by comparing the deviance for each model and also for individual data points to identify outliers. As each model has the same number of parameters, the deviance statistic could be compared directly and the model with the lower deviance selected as the best fitting. Visual fit was assessed in two ways. First, the cumulative normalized distribution of the observed and expected quantities of poultry traded by distance were plotted using Kaplan Meier methods. Second, scatter plots of the observed and expected poultry flow were plotted with the line $y=x$ used to aid the identification of outliers. Correlations between the observed and expected data were calculated using Pearson's correlation coefficient (r) in STATA v10.

7.2.3 Description of Data

Human population data: The most recently available data on province and district level human populations were obtained from a 1998 census conducted by the National Institute of Statistics in the Ministry of Planning (NIS 1999; NIS 2002). The human population sizes of the districts in the 24 study districts and Phnom Penh are presented in Figure 7-3.

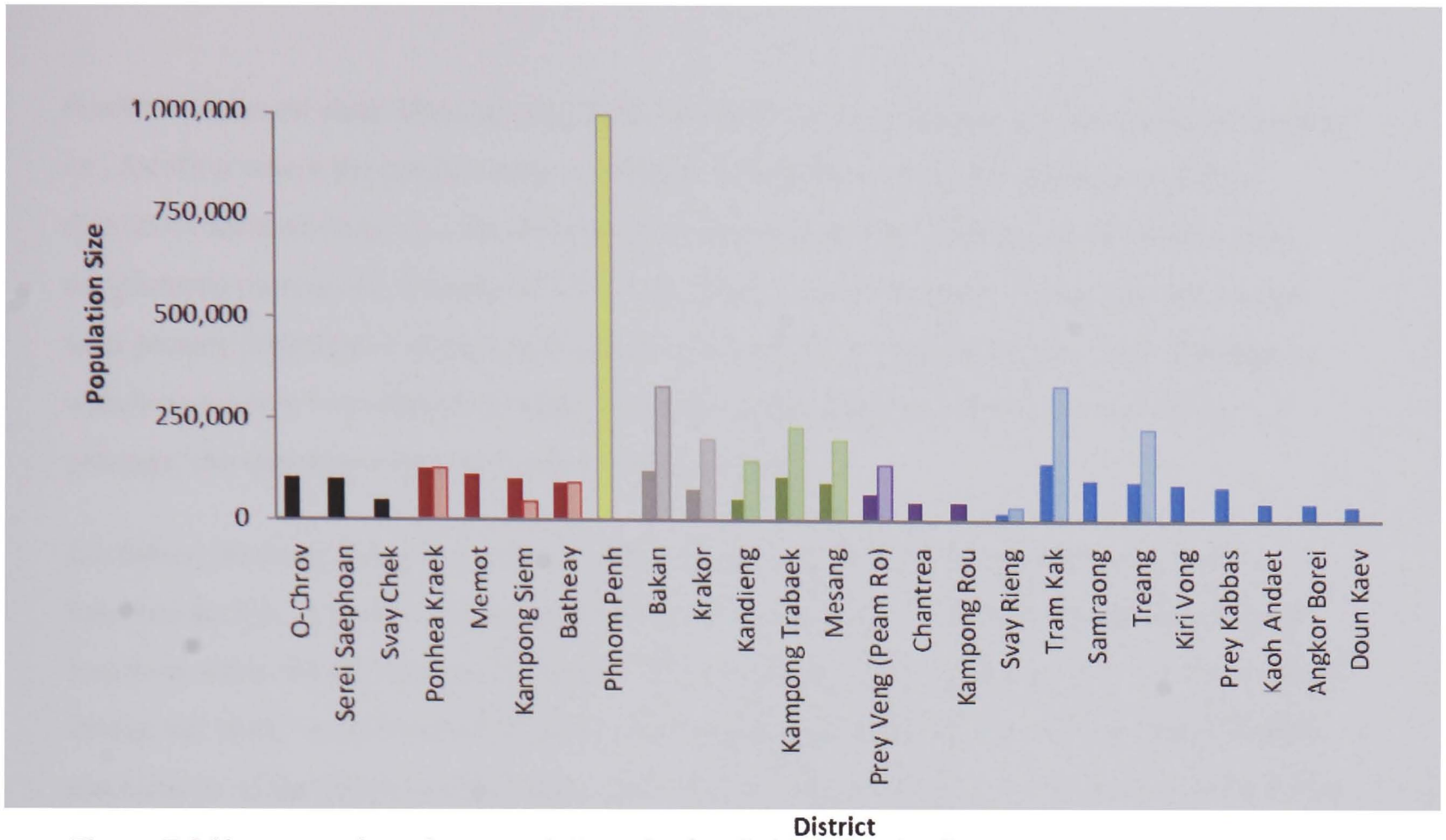


Figure 7-3 Human and poultry population size by district, Cambodia

The figure shows human population data in Black: Banteay Meanchey; Red: Kampong Cham; Gray: Pursat; Green: Prey Veng; Purple: Svay Rieng; Blue: Takeo; and poultry population data in lighter shades: Pink: Kampong Cham; Light Gray: Pursat; Light Green: Prey Veng; Light Purple: Svay Rieng; Light Blue: Takeo.

Poultry population data: Poultry population data are not available at the district level from any known source, although an estimate of the poultry (chickens and ducks) population at the province level was provided by NaVRI (MAFF Unpublished Data). Poultry populations at the district level were calculated as:

$$Poultry\ population\ at\ N_i = H_{District} N_i \times \frac{P_{Province} N_i}{H_{Province} N_i}$$

Where:

- $H_{District} N_i$ = Human population at the district level of N_i (NIS 2002)
- $P_{Province} N_i$ = Poultry population at the provincial level of N_i (MAFF Unpublished Data)
- $H_{Province} N_i$ = Human population at the provincial level of N_i (NIS 2002)

Calculated poultry population sizes of the districts included in the analyses are shown in Figure 7-3.

Poultry movement data: Data on poultry movement was reformatted into the district of origin (i i.e., location where the poultry were purchased by market sellers and middlemen) and the district of destination (j , i.e., the destination where poultry were sold by market sellers and middlemen) pairings (i, j) using STATA v10. The observed quantity of chickens, ducks and total poultry (chickens + ducks) sold between the each (i, j) pairing was recorded. Pairings in which no poultry were traded between were not included in this analysis. For identical (i, j) pairings, the quantity of poultry traded was aggregated.

Euclidean distance between locations where poultry are traded: The Euclidean distance between each (i, j) pair was calculated using GPS coordinates. The GPS coordinates of most locations were obtained during the study. GPS coordinates for locations which were not visited during the study were obtained from the 1998 Cambodian census data, and represent the GPS coordinates of the district centre (NIS 1999). The Euclidean distance between the two locations (X_i, Y_i) and (X_j, Y_j) was calculated as:

$$Distance \text{ (in kilometres)} = \sqrt{(X_j - X_i)^2 + (Y_j - Y_i)^2}$$

Calculated road distance between locations where poultry are traded: Road distances between (i, j) pairs were calculated in ArcGIS 9 using GPS coordinates, road networks and ferry crossings in Cambodia. To calculate road distances using this method, a network analyst data set using the Cambodian road network was built in ArcGIS using the GPS points of all of the locations. Using the network analyst “solve” function, the calculated network distances between each GPS point and every other GPS location were calculated to create a cross-tabulation distance matrix (ArcGIS 2006). Calculated road distances were then abstracted from the cross-tabulation distance matrix.

Travel times between locations where poultry are traded: A third measure of distance between each (i, j) pair was the estimated driving time, which takes into account road distance, road conditions, traffic, and ferry crossings. Estimates of the driving time were obtained from three drivers and one epidemiologist from Institut Pasteur Cambodia and represent driving by car. Driving time estimates via motorbikes were not obtained.

Included/excluded data: Only poultry movement data originating from the 24 included study districts (Figure 3.3) are included in this analysis since they represent a reasonably complete network in these districts, whereas poultry movement data from areas outside of the 24 study districts are incomplete. Thus, poultry originating from locations outside Cambodia (i.e., Vietnam, Thailand) are not included in these analyses. A total of 60 matched origin and destination pairs (i, j) were available for use in this analysis, however 15 of these were excluded from model fitting because $i = j$ (poultry were traded within the district and therefore distance=0). Therefore these analyses are based on 45 pairs with associated quantities of poultry traded. All 45 pairs represent unidirectional movement. There was no bi-directional movement observed (see Chapter 6).

7.3 Results

Figure 7-4 illustrates the contact networks of chicken and duck movement as shown in Chapter 6 overlaid onto human population density maps. This is a crude indication that large amounts of poultry may be traded from areas with higher human population density.



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Figure 7-4 Directed network of chicken (above) and duck (below) trading in Cambodia shown on map of human population density

The colour of the nodes represents location type (red=rural source, black=market, yellow= foreign source, green=commercial farm (CP farms), pink = semi-commercial farm) and are weighed by out-degree. The links are directional as indicated by the arrow and the tie strength is weighted by the number of birds traded between nodes (larger arrows indicate a larger quantity of birds traded between the two locations).

Among poultry traded to locations outside of the district where poultry were raised, the median distance poultry are traded is 54.8 km (IQR: 30.2-82 km; last row in Table 7-1). The median number of chickens, ducks and total poultry traded each week between any two locations is 350.4 (IQR: 80-1559.4), 0 (IQR: 16.2-249.8), and 540 (IQR: 148-1774.2), respectively. The quantity of poultry traded within districts was smaller than the poultry traded to locations outside of the home district.

Table 7-1 Summary statistics of poultry movement for 45 i, j pairs

Data Grouping: distance traded	Number of i, j pairs	Median Distance (IQR)	Median Quantity of birds traded from i, j n (IQR)		
			Chickens	Ducks	Total Poultry
Distance = 0	15	0 (--)	50.0 (14.50 156.25)	2.5 (0-23.3)	85.0 (18.8-161.5)
Distance \neq 0 km	45	54.8 (30.2-82.5)	350.4 (80-1559.4)	0 (16.2-249.8)	540 (148-1774.2)

7.3.1 Model Fitting

7.3.1.1 Model fitting using human population data

Figure 7-5 shows a gravity model fitted to the observed poultry movement using human population data and the exponential spatial kernel. Figure 7-6 shows the same model using the power law spatial kernel. Parameter estimates for these models are shown in Table 7-2.

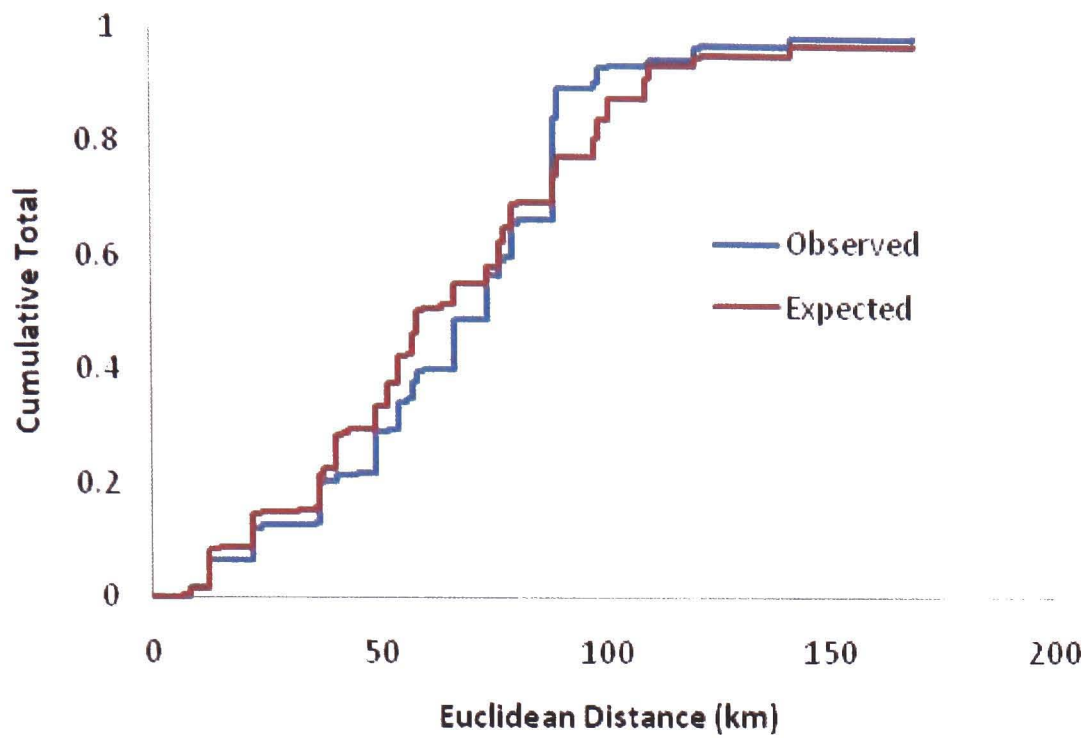


Figure 7-5 Cumulative distribution of poultry movement by distance fitted to a gravity model using human population data and an exponential spatial kernel

The plot shows the model $f(d, H_i, H_j) = H_i^\alpha H_j^\beta k^*(d_{ij})$ fitted to observed poultry movement data where distance $\neq 0$ and human population data at i and j .

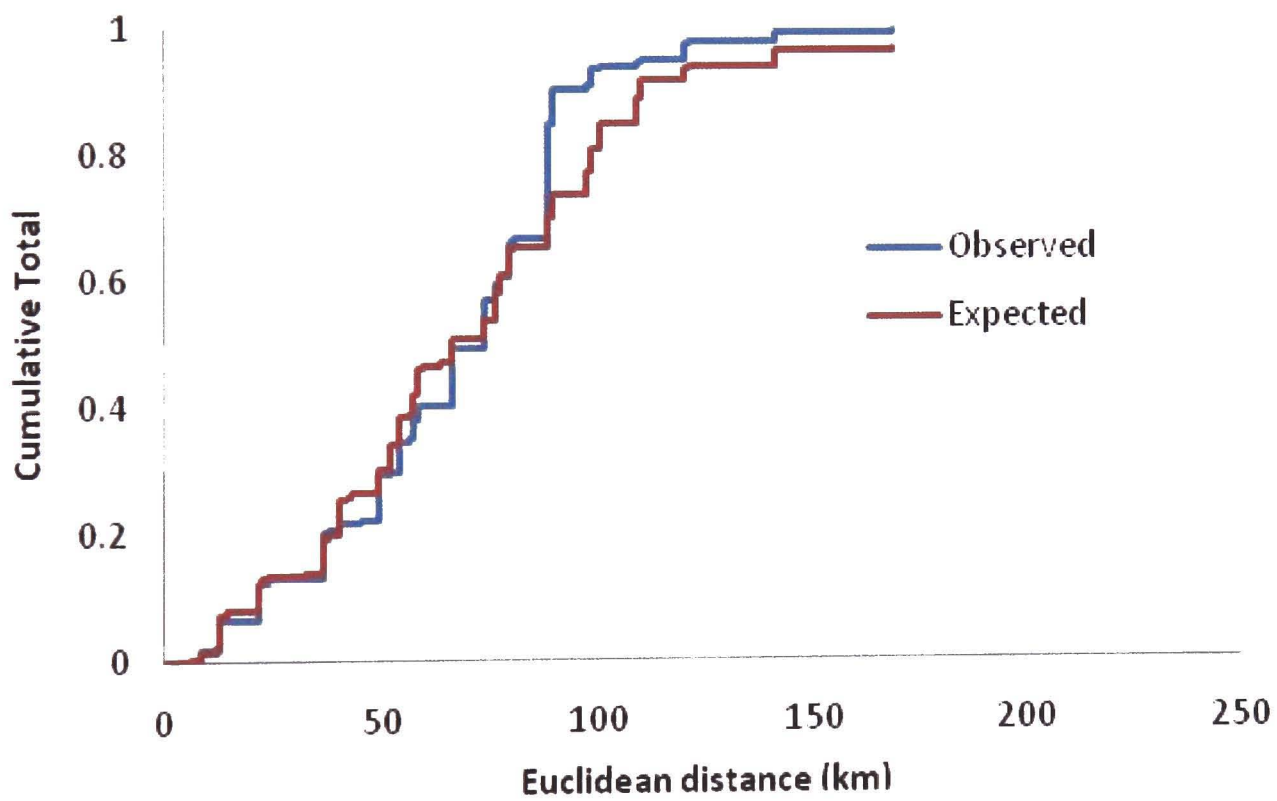


Figure 7-6 Cumulative of poultry movement by distance fitted to a model using human population data and a power law spatial kernel

The plot shows the model $f(d, H_i, H_j) = H_i^\alpha H_j^\beta k^*(d_{ij})$, fitted to observed poultry movement data where distance $\neq 0$ and human population data at i and j .

Table 7-2 Parameter estimates for models fitted to poultry movement and human population data

Parameter Estimates	Spatial Kernel	
	Model using an exponential function	Model using a power law function
G	0.1154	0.5097
Epsilon (ϵ)	0.50	0.46
Beta (β)	0.9459	0.9048
Lambda (λ)	120.0848	4052.9756
Gamma (γ)	--	31.9928
Deviance	53235.94	55146.02

The deviance is lower for the model using an exponential spatial kernel compared to the model using a power law spatial kernel indicating an overall better fit of the model using the exponential spatial kernel. Thus, the results provided below are of models fitted to data using an exponential spatial kernel.

7.3.1.2 Models Fitted to Poultry and Human Population Estimates

As the model using human population data alone did not appear to fit the data, I subsequently fitted the model using data on the poultry at the source and human population at the destination to try to improve the model fit. The results of the model fitting using poultry and human population data and an exponential spatial kernel are shown in Figure 7-7 with corresponding parameter estimates in Table 7-3. A single model appears to fit all distances and hence the model was not stratified any further.

Table 7-3 Parameter estimates for model using Euclidean distance

Parameter	Best fit estimate
G	0.0473
Beta (β)	0.9756
Epsilon (ϵ)	0.6068
Lambda (λ)	91.060
Deviance	48176.96

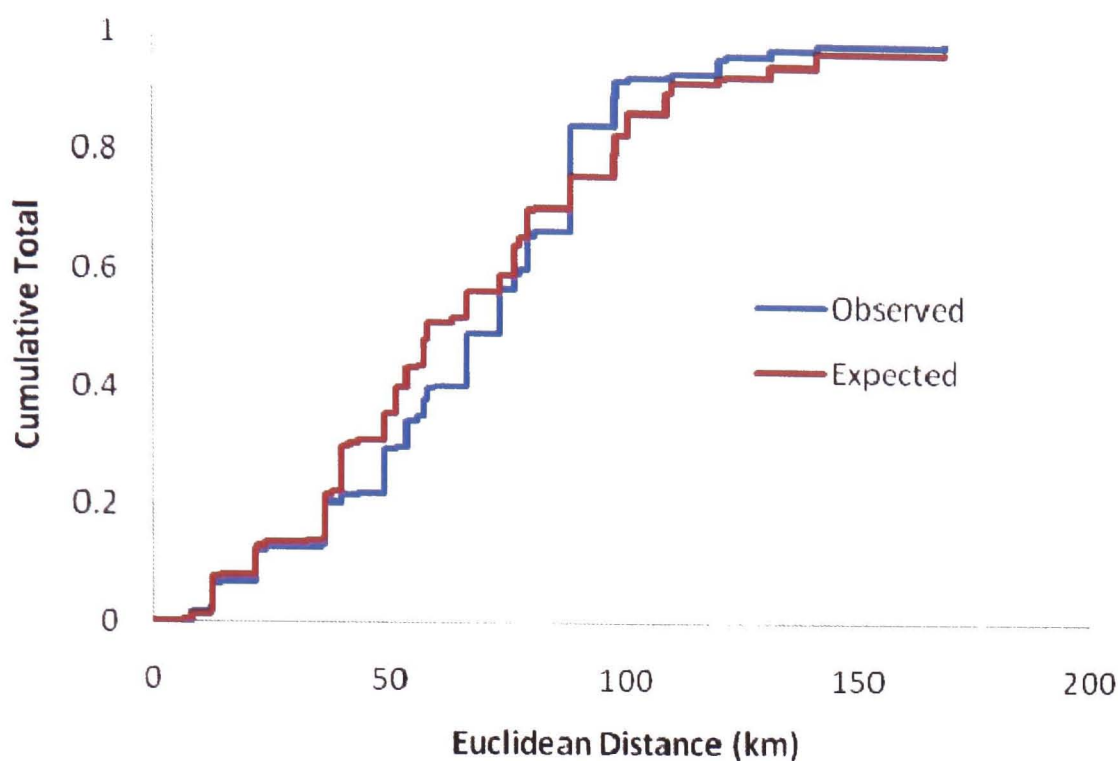


Figure 7-7 Cumulative distribution of poultry movement by Euclidean distance

The plot shows the model $f(d, P_1, H_2) = GP_1^\epsilon H_2^\beta k^*(d_{ij})$ fitted to all data where distance $\neq 0$

From Figure 7-7 it is noticeable that there is a divergence between the observed and model predicted distributions which begins at approximately 50-75 km where the observed number of poultry traded at these long distances is less than predicted by the model and again at distances greater than approximately 100 km where the observed number of poultry traded is larger than predicted by the model. One possible reason for this may be that poultry traded at long distances follows a different economic model to that traded at short distances. For example, one hypothesis would be that at short distances most poultry can be traded by middlemen on motorbikes but at longer distances this is no longer feasible and thus fewer poultry are traded because of a relative lack of availability of trucks.

7.3.1.3 Model Fitting Using Calculated Road Distances

Using Euclidean distances has many limitations particularly in Cambodia where there are relatively few good quality roads. The same analysis was therefore performed using calculated road distances as a more relevant measure of the trading distance between two locations.

There is a strong correlation between Euclidean distances and calculated road distances ($r=0.89$) although there is a tendency for calculated road distances to be longer than Euclidean distances (Figure 7-8).

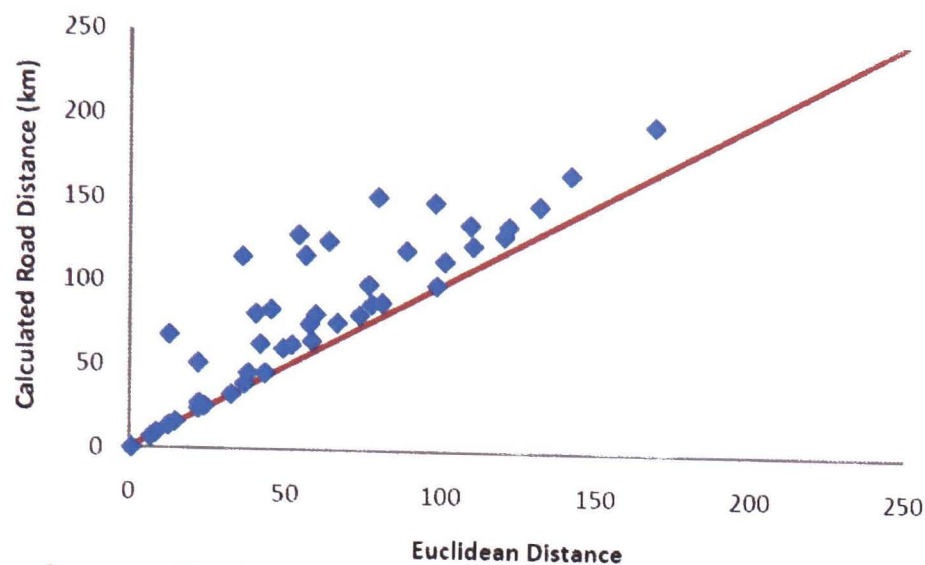


Figure 7-8 Euclidean distance (km) vs. calculated road distance (km)

A model was fit using data on the poultry at the source, human population at the destination, calculated road distances between i and j and an exponential spatial kernel (Figure 7-9). A single model appears to fit all distances and hence the model was not stratified any further. Parameter estimates of the model are provided in Table 7-4.

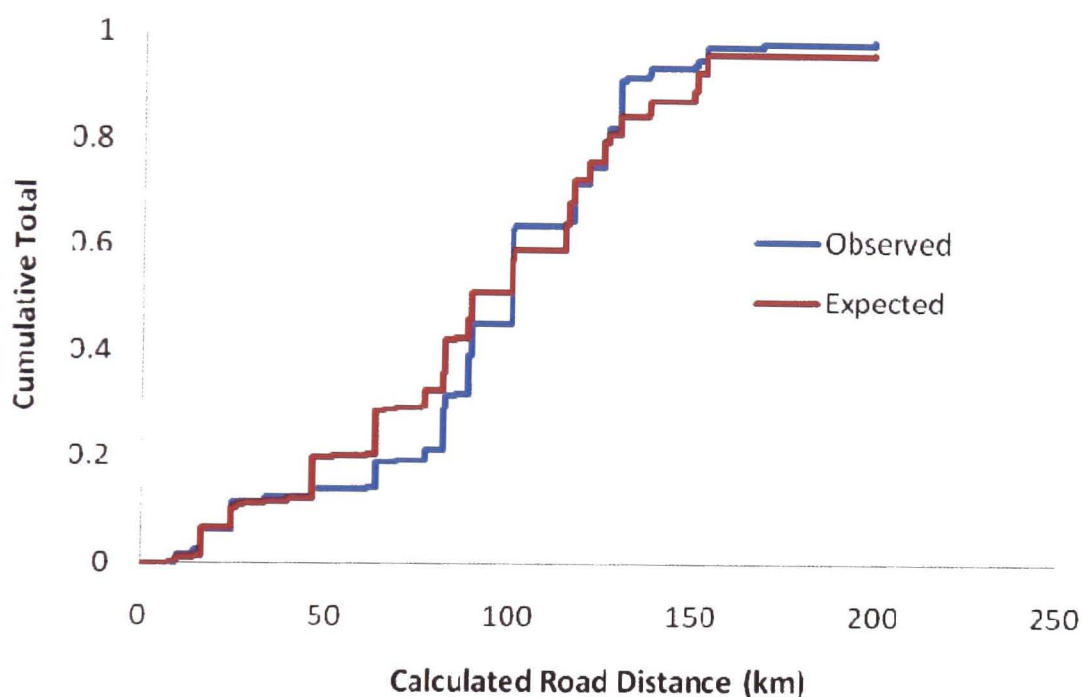


Figure 7-9 Cumulative distribution of poultry movement using calculated road distances

The plot shows the model $f(d, P_1, H_2) = GP_i^\epsilon H_j^\beta k^*(d_{ij})$ fitted to all data where calculated road distance $\neq 0$

From Figure 7-9 the same divergence is seen between the observed and predicted poultry flow at distances between 50-100 km and above 125 km indicating that that poultry traded at long distances, this time above 125 km follows a different economic model to that traded at short distances (<125 km).

Table 7-4 Parameter estimates for model using calculated road distances

Parameter	Best fit estimate
G	0.5002
Beta (β)	0.9339
Epsilon (ϵ)	0.5427
Lambda (λ)	167.4561
Likelihood ratio statistic	53293.8044

7.3.1.4 Model Fitted to Distance Measured as Journey Time

The estimates for the journey time were strongly correlated with Euclidean distances ($r=0.7699$; Figure 7-10 left), although this correlation was not as strong as that between calculated road distance and time ($r=0.89$; Figure 7-10 right).

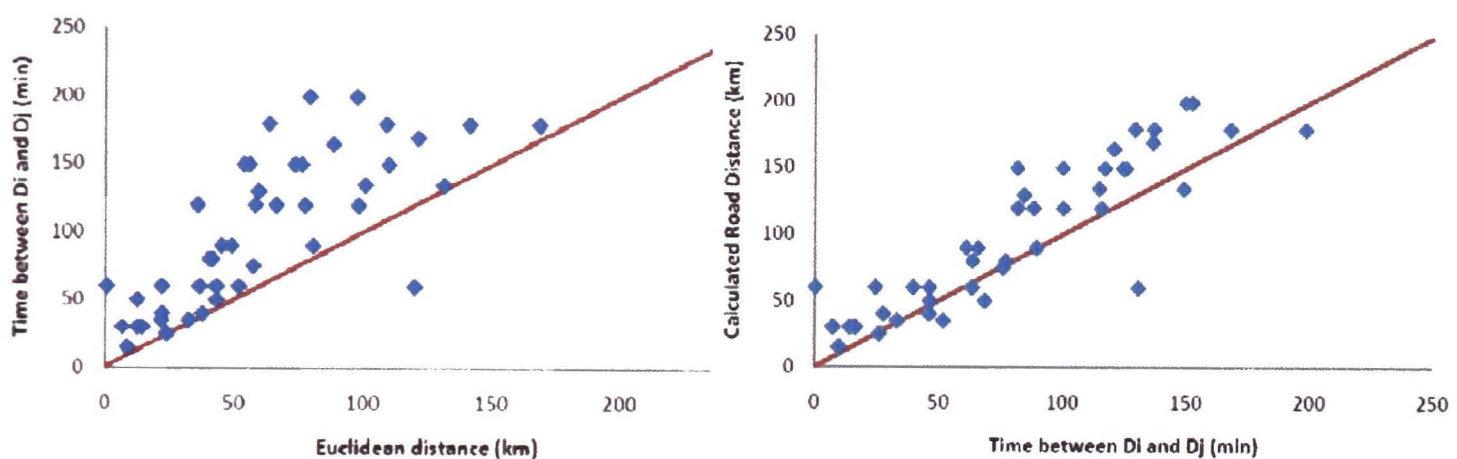


Figure 7-10 Euclidean distance (km) vs. Time to travel between i and j (min; left) and Calculated road distance (km) vs. Time to travel between i and j (min; right)

The results of the model fitted using travel times as a measure of distance is shown in Figure 7-11 with corresponding parameter estimates for the model in Table 7-5. A single model appears to fit all distances and hence the model was not stratified any further.

Table 7-5 Parameter estimates for model using journey time between i and j

Parameter	Best fit estimate
G	2.6839
Beta (β)	0.9240
Epsilon (ϵ)	0.46670
Lambda (λ)	240.3828
Deviance	54534.09

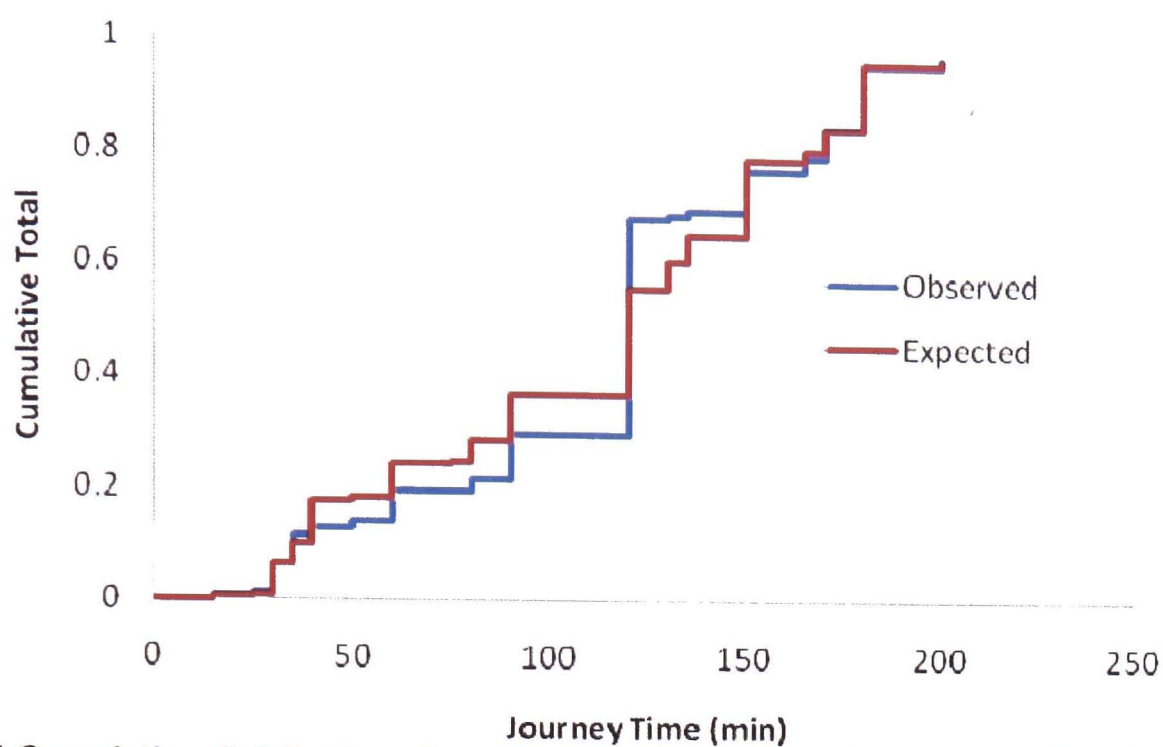


Figure 7-11 Cumulative distribution of poultry movement using distance as a function of time

7.3.2 Evaluation of the model fits

The parameter estimates and deviance for all three models using the three measures of distance (Euclidean distance, calculated road distance and journey time) are shown in Table 7-6 and the fitted spatial parameters for all three models are shown in Figure 7-12.

Table 7-6 Parameter estimates for models using Euclidean distance, calculated road distance and journey time

Parameter	Estimates for models using:			
	Human Population Data and Exponential Spatial Kernel	Euclidean Distance	Calculated Road Distance	Journey Time Distance
G	0.1154	0.0473	0.5002	2.6839
Beta (β)	0.9459	0.9756	0.9339	0.9240
Epsilon (ϵ)	0.50	0.6068	0.5427	0.4670
Lambda (λ)	120.0848	91.0604	167.4561	240.3828
Deviance	53235.94	48176.96	53293.80	54534.09

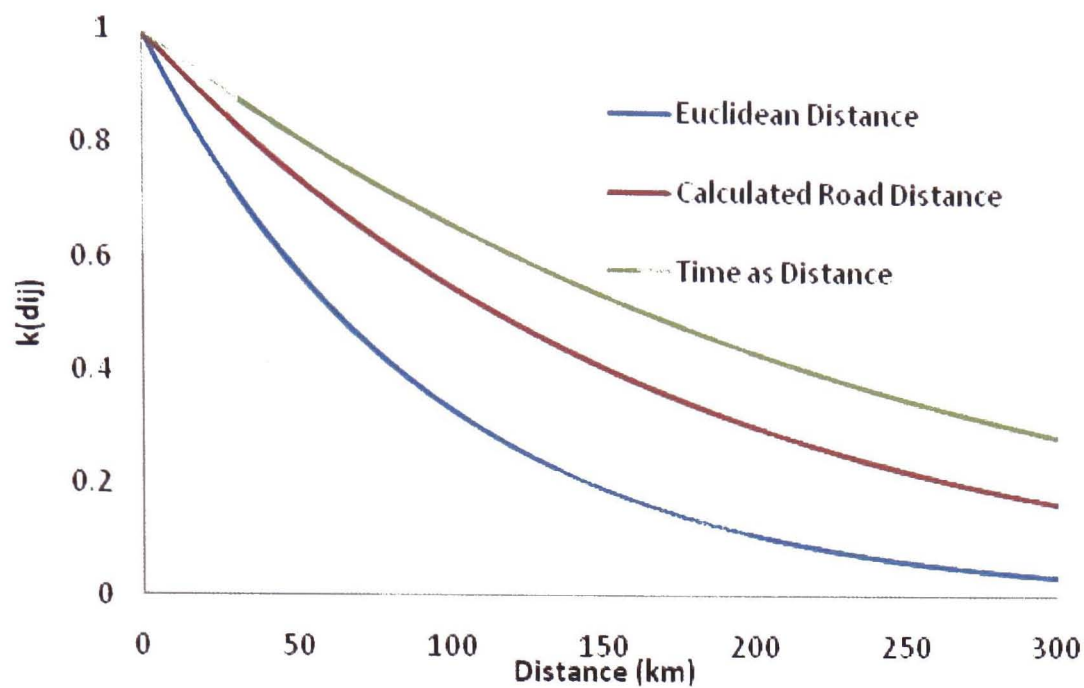


Figure 7-12 Fitted spatial kernel parameters plotted for models using Euclidean distance, calculated road distance and journey time

The model using the Euclidean distance best fits to the observed poultry movement data (lowest deviance of all the models), while there was little difference in the fitted spatial kernels of the three models indicating that the quantity of poultry traded rapidly declines with distance. To further evaluate the fit of the model the observed quantities of poultry flow are plotted against the model predicted quantities for all poultry movement and an $y=x$ line (Figure 7-13). There is a strong correlation between the observed and predicted data ($r= 0.61$) indicating that a reasonably high degree of variation in the data is being explained by the model. However, a number of outliers are noticeable in this plot.

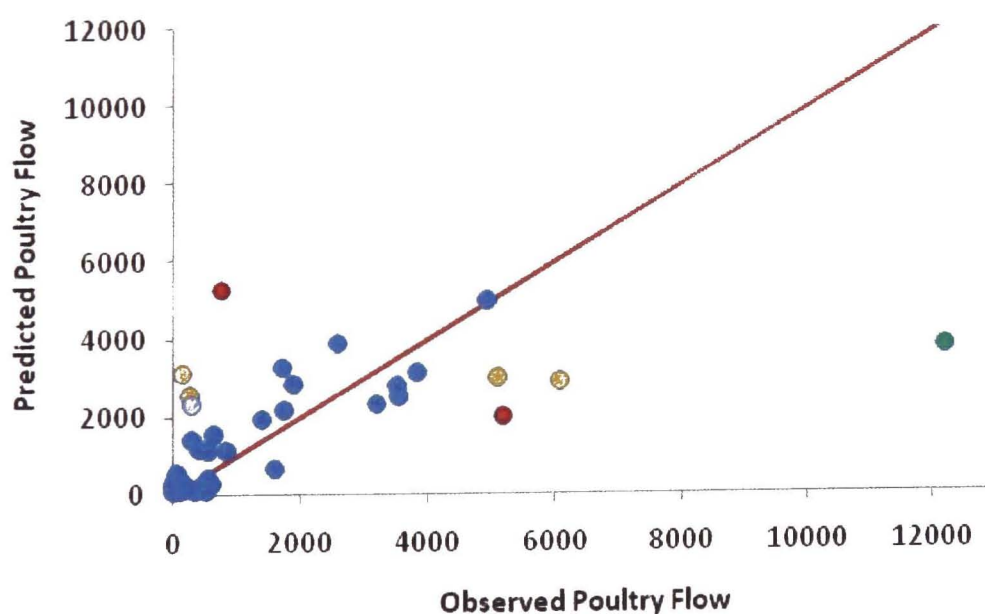


Figure 7-13 Observed vs. predicted poultry flow between $i - j$ using Euclidean distance

This plot shows the observed vs. predicted poultry flow along with a $y=x$ line.

All of the data points highlighted in orange and grey represent poultry movement into Phnom Penh from Prey Veng (orange, n=4) and Svay Rieng (grey, n=1) provinces. All journeys from these provinces must cross the Mekong River at a ferry crossing at Neak Leung, Prey Veng to reach Phnom Penh, which can add up to one hour wait time to any journey crossing the river. Thus, this may explain why the number of poultry predicted by the model is higher than that observed (i.e., 3 nodes above the $y=x$ line). However this does not explain why the number of poultry predicted by the model is lower than observed (2 nodes below the $y=x$ line).

The data points highlighted in red and green represent poultry movement from Takeo province into Phnom Penh. The location identified in green represents poultry trading from a large poultry market in Tram Kak, Takeo along National Road 3 into Phnom Penh with over 12188 birds (76.6% chickens, 23.4% ducks) traded weekly. There may be large quantities of poultry from Kampot province, which borders this area of Takeo, that are traded at this market and onto Phnom Penh via middlemen, although market sellers and middlemen purchasing poultry from this area did not indicate this. This is clearly an outlier in the dataset. However the parameter values did not change when removing this data point from the analysis.

Figure 7-14 shows the corresponding data using a log scale and colouring the data points for poultry movement into Phnom Penh (blue) and localized poultry movement to and from locations outside of Phnom Penh (yellow).

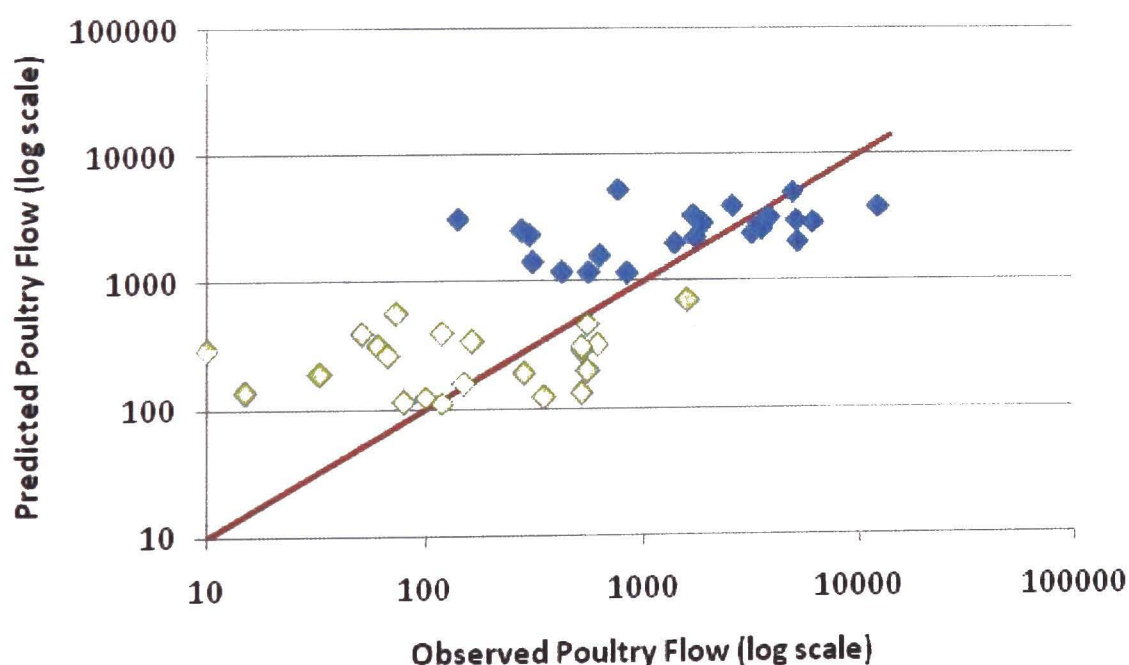


Figure 7-14 Observed vs. predicted poultry flow using a log scale for the model using Euclidean distances

The data points in yellow represent poultry trading in rural regions of the country (e.g., within Banteay Meanchey province, within Pursat province, within Svay Rieng province and within Takeo Province). There was less variation in the predicted poultry flow for localized poultry trading and poultry movement into Phnom Penh than was observed. The model appears to under-estimate the amount of poultry flow occurring for approximately half of these locations ($n=9/22$) while over-estimating the amount of poultry flow for the other half of these locations ($n=11/22$). A similar pattern of predicted flow is seen for trading to locations in Phnom Penh. However, there were no improvements seen in the model fit after adjusting for trading to locations in Phnom Penh indicating that there are other cost factors that are affecting locally traded poultry versus poultry traded to locations in Phnom Penh.

7.4 Discussion

These results illustrate that poultry movement is best described using poultry populations at the source (i.e., the supply of poultry) and human population at the destination (i.e., the demand for poultry). Regardless of the distance measure used in the model fitting, the parameter estimates for the poultry population (ϵ) and human population (β) in the model remained relatively constant. Epsilon (ϵ) ranged from 0.47 – 0.61 and β ranged from 0.92 – 0.98 under the different models. Epsilon is a parameter which scales the influence of the source population and if equal to 0 would indicate that every source location (i.e., district in this dataset) sells a constant number of chickens and ducks regardless of poultry population size. If $\epsilon=1$ then poultry are sold proportionately to the poultry population size of the source location, whereas if $\epsilon>1$, this would indicate that relatively more poultry are sold from districts with larger poultry population sizes than from districts with smaller poultry population sizes. An $\epsilon>1$ may signify that these areas may keep poultry to sell as part of their livelihood, which from my studies has been shown to rarely occur in rural areas of the country (Chapter 6). However, for all of the models, the parameter estimates for ϵ were less than one which may indicate that relatively fewer poultry are being sold from districts with larger poultry population sizes than from districts with smaller poultry population sizes. This may suggest that individuals in rural areas are only able to sell poultry when they have a surplus, a situation which rarely occurs in rural Cambodian households (Chapter 6).

Beta is a parameter which scales the influence of the destination population (i.e., demand). A $\beta > 1$ would indicate that large human populations may be buying proportionately more poultry than small populations, which is what one would expect if larger populations are located in more urban areas and therefore there is a greater demand for poultry from individuals living in more densely populated areas as compared to rural areas. All of the β parameter estimates for the models were close to 1 indicating the human populations at the destination are buying proportional to the size of their population.

Presentation of the model fit using scatter plots was an informative tool to evaluate the fit of each model and was especially useful in identifying possible outliers. It would not have been possible to identify possible outliers if I had only produced the cumulative distance distribution graphs for each model. Local knowledge of the poultry trading system in Cambodia has been helpful in interpreting these results, especially with respect to the characteristics of the outliers.

As was expected, these results show that there is not a linear relationship between poultry quantity traded and distance travelled, and the models were sensitive to instances where large quantities of poultry are traded short distances (e.g., 3500 poultry traded <20 km) and conversely when small quantities of poultry are traded over long distances (e.g., 281 birds traded >130 km; 309 birds traded approximately 170 km). Within Cambodia, it is not uncommon for large quantities of poultry to be traded over short distances because there were several large markets located within 20 km from Phnom Penh. Therefore there may be a different economic model for poultry traded locally versus traded into Phnom Penh.

The models seem to suggest that there may be different cost factors for localized poultry movement, i.e., poultry traded to locations outside of the home district but within the same province of origin than for poultry traded into markets and other locations in Phnom Penh. Possible explanations for this trend may be that there are different economic drivers for poultry traded for longer distances as compared to shorter distances or that the mode of transportation (e.g., motorbike vs. truck/van) makes longer distances possible for some middlemen. It is possible that separate models are required for shorter and longer poultry movements, however because my data covered poultry movements over a relatively short distance (up to 200 km), I saw no evidence using the deviance statistic of a better fit in the models by distance and did not

find evidence of a better fit when categorizing poultry trading at different distance thresholds (e.g., <75 km vs. \geq 75 km using Euclidean distance; <125 km vs. \geq 125 km using calculated road distance; data not shown). A measure of the mode of transportation (e.g., motorbike, truck, bicycle, foot) would be useful to incorporate into the analysis to evaluate the fit of the models and changes in parameter estimates (e.g., $\beta >$ than 1 for poultry traded to locations in Phnom Penh and $\beta <$ 1 for localized trading).

Using the Euclidean distance provided models with the best fit to the observed data, which was an unexpected finding since calculated road distances are a more exact measure of the distances travelled between each location. Although the values for calculated road distances were highly correlated with Euclidean distance and also with journey time, there may have been some inaccuracies in the values obtained for calculated road distance and journey time. For example for the calculated road distances, the road network in ArcGIS did not originally include any bridged or ferry crossings and in order to obtain a more accurate calculation of road distances I provided an approximate location of the ferry crossing at Neak Leung and an important bridge linking National Road 1 and Phnom Penh. Without this information, calculated road distances from locations in Prey Veng and Svay Rieng were 3-4 times larger than they should have been as compared to road distances calculated by the Ministry of Public Works and Transport (MPWT 2009).

Although calculated distances are a more accurate representation of the distances travelled, they still did not capture the “costs” associated with each journey. In using a measure of the time to make the journey between each location, the measure is able to take into account the road conditions, which if they are made of dirt are virtually impassable for several months out of the year; traffic, which in some areas leading into Phnom Penh can delay a journey for 15-60 minutes depending on the time of day; and the use of a ferry to cross the Mekong River, which is required for all journeys originating in Prey Veng, and Svay Rieng Province (and including Vietnam). Ideally this measure should include the vehicle type (e.g., car, truck, motorbike, bicycle) if it were to truly capture the journey times made by poultry traders in this region. However, there was no evidence of a better fit of the model when using journey time as

the distance measure, which may indicate that there are inaccuracies with the estimates I obtained.

Some movements are not well captured by any of the models, specifically poultry traded into Phnom Penh and some poultry traded to Peam Ro which is the ferry crossing at Neak Leung in Prey Veng Province. This may be because these models fitted to poultry movement data in Cambodia do not take into consideration the costs associated with trading poultry. For example, the cost of diesel may prohibit some traders from making longer journeys into Phnom Penh especially if they are carrying only a small number of live birds on their motorbikes each week thus limiting the profit they are able to make with each journey. On the other hand, if middlemen use trucks for transportation, they can carry thousands of live chickens and ducks each week, resulting in the marginal cost of diesel compared to the profit made by the sale of their poultry. There are often multiple middlemen transporting poultry on the same truck, which would further reduce the cost of diesel if they were dividing the cost. An estimate of how poultry are traded for each journey may explain some of the variation in the poultry flows predicted by the model.

Since the poultry network is highly centralized, a large proportion of the locations where poultry are sold are in Phnom Penh (24 of the 45 pairs), where the population size is larger by a factor of ten compared to districts in rural areas. Because the population sizes for all identical destinations are the same, there may be other factors not accounted for in the model that are driving poultry movement. Again, an estimate of the cost/profit made from poultry sold locally as compared to poultry sold to locations in Phnom Penh would be important to incorporate into the model. However, I did not collect this information from my subjects and I am unaware of any existing data that could address these issues.

These analyses were limited by the use of districts as the origin and destination rather than the actual location where poultry were purchased and sold (e.g., village or market). This was decided because I had GPS coordinates for all nodes either obtained during visits to the actual locations or from GIS software. When GIS software was required, the district centre was used instead of the actual location. Although the included districts are not large, this may have

impacted the results since district centres tend to be located along paved roads whereas villages and some smaller poultry markets may be located along dirt roads.

The use of disaggregated data of poultry traded between identical locations is warranted to determine if identical journeys that are made by multiple individuals shed light on other factors driving poultry trade. It is unclear what effect these differences may have had on the results; however further analyses are necessary since there were only a small number of such pairs available for model fitting (n=45).

Another limitation of this analysis is that I only considered pairs of locations in which movement was observed to occur (n=24 districts) and therefore, the current models did not capture the lack of movements to other locations nor the movements between districts that were not sampled (n=159 districts). This may result in biased estimates of the model parameters. For example, if there were a nearby district with a large human population to which poultry did not move, including this would have the effect of reducing the estimate of the power parameter on the human population. Future analyses will incorporate this additional information. However, in order to do so, it will be necessary to condition the analysis (using a multinomial rather than Poisson likelihood) on the number of poultry observed to leave the study populations as the survey did not fully capture movements from other locations. However, this would result in a model that no longer predicts the factors driving the source population (i.e. the number of poultry at the source population) which limits the application of the model to other settings with different underlying poultry population distributions.

These analyses are also limited by estimates of human and poultry populations that are unable to be verified. The last census conducted in Cambodia was conducted in 1998 and therefore do not take into account growth rates, which vary disproportionately throughout the country (CIA 2008), nor do they take into account changes in rural/urban living. In addition, there was no way for me to assess the quality of estimates of poultry population at the provincial level since the methods by which these values were collected were not provided by NaVRI. A more accurate estimate of poultry populations at the district level are needed.

Despite these limitations, using gravity models has been useful to uncover relationships between poultry movement, distance and human/poultry populations at the source and destination. These results have demonstrated that gravity model theory can be applied to poultry movement data. This is a novel application of the use of gravity model theory as there are no studies that have fitted gravity models to animal movement data.

Once validated, these results may be able to predict trade flows not covered by the study areas and elucidate poultry movements within the Mekong Delta Region. Since it is not feasible to collect the movement of poultry for the entire Mekong Delta Region, gravity model theory is a useful tool to predict poultry movement in a wider area to gain insight on how these movements could be controlled to prevent the spread of HPAI.

Chapter 8 Conclusions and Future Work

8.1 Key Findings

My PhD research involved the collection of novel and original data from rural Cambodians, rural, peri-urban and urban market sellers and poultry middlemen from six provinces and Phnom Penh between April 2006 and December 2007. In my first large-scale survey of randomly selected adults and children living in rural areas of Cambodia, I evaluated poultry ownership and husbandry practices, poultry mortality experienced and poultry mortality reporting. This study also evaluated the extent and frequency of poultry handling behaviours of each subject and how these practices differ by age and gender. This is the first study to evaluate poultry contact patterns at an individual level and the first to evaluate poultry contact patterns of children. My second study identified and interviewed rural, peri-urban and urban poultry market sellers and middlemen to evaluate their poultry trading patterns. Through these two studies, this thesis has evaluated poultry movement and the extent of interaction between humans and poultry in Cambodia to better understand the risks of sustained transmission of H5N1 in poultry and onward potential transmission to humans.

8.1.1 HPAI and Poultry Populations

The results from my first study demonstrated that most rural Cambodians own small quantities of chickens and/or ducks. Although a large majority of backyard flocks generally consist of less than a dozen chickens, most domestic poultry often mix with other domestic animals, including pigs, water buffalo, and dogs. Thus there is the potential for genetic mixing of an avian and non-avian influenza virus in these species (Alexander & Brown 2000; Horimoto & Kawaoka 2001).

The use of bio-security on farms with backyard poultry was found to be minimal or non-existent and could be readily improved to a minimum level by keeping poultry species separated from other domestic species, separate from human areas, and restricted to forage for food within a specified and controlled area. One way to reduce the proportion of free-ranging animals and thus the potential for HPAI transmission between flocks is to improve the sealant

properties of fences and netting that are used by approximately 25% of households in rural areas of the country. However, feasible options for providing feed to enclosed flocks would be necessary as flocks would no longer be allowed to freely forage for food. This presents a major challenge to HPAI control in developing countries.

My research has highlighted the heterogeneity in poultry raising practices that would normally be classified as Sector 4 Poultry Production according to FAO definitions (FAO 2006). In order to have more transparent and more informative descriptions of poultry raising in countries with predominant Sector 4 holdings and to design effective control measures for HPAI, further information is required, such as the size of poultry flocks, the species raised, and the output into the poultry market chain.

In Cambodia, more than 90% of backyard flocks are composed of small numbers of free-ranging chickens (Sector 4, Subcategory A). However among households that raise poultry in Cambodia, approximately 1% of households included in my study rear ducks ($n > 50$) using a minimal level of bio-security (birds are kept indoors or allowed to be free-ranging for part of the day and kept indoors or fenced in at night; Sector 4, Subcategory B), whereas approximately 6% of households raise small numbers of free ranging ducks (Sector 4, Subcategory B). These two subcategories represent the greatest potential for sustaining H5N1 circulation in rural Cambodia since infected ducks can silently spread the virus (Chen et al. 2004; Hulse-Post et al. 2005). This is supported by research from Thailand, which has shown that free-grazing ducks were strongly associated with HPAI outbreaks in villages and thus were crucial for the persistence and spread of HPAI in Thailand in 2004 (Gilbert et al. 2006; Gilbert et al. 2007). Thus, it is especially important for HPAI surveillance to monitor the HPAI status of Sector 4, Subcategories A and B holdings since they represent the greatest HPAI risk. Control efforts in Thailand that focused on regulating movement and monitoring H5N1 infection via serologic and virologic testing of duck flocks may have led to the reduction of H5N1 outbreaks in 2006 (Webster et al. 2007).

My research has also revealed that it is not uncommon for households to experience poultry mortality, with more than half of the households experiencing some degree of poultry mortality within the previous 8-month period. A key finding is that respondents reported an average of

50% within-flock mortality among both chicken and duck flocks over the study's recall period. Of cause for concern is that this level of mortality was not normally treated as suspect by villagers, even when within-flock mortality exceeded 50%, 60% or 75%. It is probable that there has been considerable underreporting of HPAI in poultry populations in rural areas of the country and therefore there remains a strong need to improve the passive HPAI surveillance system in Cambodia. The current system relies on village animal health workers (VAHW), who have been trained by FAO and NaVRI, to identify suspect poultry mortality. Given that my study population reported difficulties in distinguishing symptoms of AI from other poultry diseases including Newcastle disease, VAHW would provide a necessary link between rural subjects and NaVRI by providing information to the subject on AI and determining whether it is necessary to report the mortality event onwards to officials at NaVRI for follow-up. Without the VAHW, it would be increasingly difficult for subjects to know where or how to report poultry mortality.

It would be prudent to focus passive HPAI surveillance on high mortality events above a specified within-flock mortality threshold. For example setting a threshold of within-flock mortality at >60% in chicken flocks or at >30% in duck flocks would help officials differentiate common poultry mortality from suspect mortality events. Using data from this study, if a threshold of 60% within-flock mortality were assumed to be the basis for which VAHW called upon ministry officials to investigate, 38 households should have been visited within the 8-month study period to investigate 30 occurrences of >60% mortality within a chicken flock and 8 occurrences of >30% mortality within a duck flock events. Given the current limited personnel and financial resources of NaVRI, investigating 38 occurrences of suspect mortality that occurred in two provinces within an 8-month period would be difficult to achieve, even more unattainable would be to investigate suspect mortality events at the national scale.

8.1.1.1 Poultry movement and HPAI

My second study evaluated poultry movement via middlemen and market sellers and illustrated that networks of poultry movement via trading in Cambodia are highly centralised, connected and unidirectional. Most poultry movement occurs into Phnom Penh making the markets in

Phnom Penh a potential hub for the spread of H5N1 and ideal for surveillance and control. Research has shown that live bird markets are an important reservoir for HPAI and an ideal environment for reassortment and transmission of HPAI from poultry-to-humans (Amonsin et al. 2008; Kung et al. 2003a; Kung et al. 2007; Nguyen et al. 2005; Wang et al. 2006; Woo et al. 2006). Thus, the movement of poultry through these markets are potentially important in the circulation and spread of HPAI (Kung et al. 2007; Sims et al. 2003). Domestic poultry outbreaks of H5N1 have occurred in areas of the main network and therefore Phnom Penh markets, namely wet markets, would be ideal for routine surveillance activities and control interventions.

Illegal cross-border trading of live poultry was also identified in this study, namely from Vietnam and to a lesser extent from Thailand. Live day-old ducks from Vietnam are directly traded with influential locations in Phnom Penh, including markets and semi-commercial farms, and make up approximately half of the total number of ducks traded weekly in Cambodia. Live and prepared poultry (birds that have been slaughtered, boiled, and defeathered) from Thailand are traded to markets in Banteay Meanchey province, but these movements are separated from the main network and I did not identify any direct links between Thailand and Phnom Penh.

Based on the results of this study, targeted surveillance recommendations were developed for NaVRI to improve their active HPAI surveillance activities. Two tiers of recommendations were developed to 1) monitor the HPAI status of poultry populations in rural areas (Tier 1 recommendations), and 2) identify locations that would allow for the early detection of HPAI incursion in markets which have a high potential for spread throughout the network (Tier 2 recommendations, Table 6-7).

The purpose of the Tier 1 recommended locations for surveillance is to directly identify locations where HPAI can be rapidly detected if the virus is in the market system and would indirectly allow NaVRI to monitor the HPAI status of poultry populations in rural areas. These recommended locations include those with the highest in-degree values, and include markets, semi-commercial farms and stock houses in Phnom Penh. If HPAI is identified in any of the Tier 1 recommended locations, an investigation to trace-back poultry back to its origin should be undertaken.

The purpose of the Tier 2 recommended locations is to identify locations that are most likely to interrupt poultry-to-poultry transmission of HPAI. The active surveillance of the locations that fall within Tier 2 would allow for the early detection of HPAI incursion in markets that have a high potential for spread. The Tier 2 recommended locations are those with the greatest number of connections and the largest out-degree scores. If HPAI is identified in any of the Tier 2 recommended locations, an investigation to trace-back and trace-forward poultry should be undertaken. This would require minimal effort to trace poultry forward as most are sold to markets in Phnom Penh and a moderate effort to trace poultry back to its origin as most are sold from nearby villages and markets.

An important finding of my second study, particularly when considering transmission dynamics of HPAI across the networks, is that poultry rarely spend more than 8 hours in the market chain before they are slaughtered. This reduces the likelihood that active HPAI surveillance activities will capture an infected or diseased bird in the markets since poultry traders did not report selling visibly sick poultry. The wet markets in Cambodia provide an ample environment for HPAI viruses to persist (Amonsin et al. 2008; Kung et al. 2003a; Kung et al. 2007; Nguyen et al. 2005; Wang et al. 2006; Woo et al. 2006) and therefore the market environment should be included in the routine HPAI monitoring activities. This is supported by an investigation of live bird markets in China where investigators found no positive H5N1 samples from cloacal swabs of several bird species from several markets, but found one positive environmental sample from a goose cage (Wang et al. 2006). Therefore, further it would be prudent for NaVRI to consider including monthly disinfection of wet market selling areas (e.g., stalls, selling platforms, locations where poultry are slaughtered) at the three main live poultry selling markets in Phnom Penh (Orussey, Chba Ampov, and Deum Kor markets) in their HPAI control strategies.

A gravity model was fit to poultry movement data using population data as an indicator of potential trade between the source where poultry are reared and destination of where poultry are sold to attempt to understand the potential driving forces behind the poultry movement patterns observed. These results illustrated that poultry movement is best described using poultry populations at the source (representing the supply of poultry) and human population at the destination (representing the demand for poultry). The models also suggest that there may

be different cost factors for poultry movement at different distances. A possible explanation for this trend may be that there are different economic drivers (e.g., mode of transportation, cost of diesel, profits from sale of poultry, journey time) for poultry traded over greater distances (poultry traded into Phnom Penh) as compared to shorter distances (localised poultry trading). This is a useful tool to predict poultry movement in wider areas and once validated, could be used to gain insight on how these movements could be controlled to prevent the spread of HPAI.

Recent unpublished evidence (presented at the Cambodia Workshop on Avian Influenza Research Activities, Sihanoukville, Cambodia 8-10 October 2008) from NaVRI's active HPAI surveillance programme, which is currently being conducted in 13 markets has revealed that H5N1 may have been circulating in markets in 2007-2008. Influenza A viruses were isolated from cloacal and tracheal swabs of birds in Phnom Penh markets in 2007 (RT-PCR influenza type A positive, H5 negative) and from birds in markets in Phnom Penh, Takeo, Pursat, Sihanoukville, Banteay Meanchey and Siem Reap (RT-PCR influenza type A positive, H5 negative) in 2008. Also in 2008, influenza A viruses, subtype H5 (RT-PCR influenza type A positive, H5 positive) were isolated from cloacal and tracheal swabs of birds from a market in Svay Rieng.

In 2007, antibodies for H5N1 were identified in poultry from markets in Phnom Penh (2/434 samples) and Takeo (16/166 samples) and in 2008, in poultry from markets in Phnom Penh (82/1155 samples), Takeo (59/390 samples), Kampong Cham (26/330 samples), Pursat (10/497 samples), Battambang (16/435 samples), Siem Reap (29/450 samples), Kampot (59/232 samples), Kandal (18/130 samples) and Sihanoukville (26/240 samples). Overall H5N1 antibodies were detected in 1.7% (18/1065 samples) of poultry tested from seven markets in 2007 and in 7.9% (325/4124 samples) poultry tested from 13 markets in 2008. These results indicate that H5N1 is circulating in the Cambodian market system. This supports the assumption that H5N1 infection has gone largely unreported in rural regions of Cambodia.

8.1.2 HPAI Transmission Risks at the Human/Animal Interface

The results from my large-scale cross-sectional survey demonstrated that most of the population in rural Cambodia is in frequent contact with domestic poultry. About half of the rural population sampled carried out, on a regular basis, at least one of the practices considered to be high risk for effective transmission if the bird is infected (e.g., slaughtering poultry, sharing water with fighting cocks, blowing into the beak of fighting cocks).

There was substantial variation in the frequency of different practices and thus the potential risk of transmission of H5N1 from poultry-to-humans is not uniform across age and gender even amongst populations living in close proximity to poultry. In conducting a semi-quantitative risk assessment of the transmission potential of H5N1 from poultry-to-humans among rural Cambodians, I determined that males between the ages of 26-40, followed closely by males between the ages of 16-25 reported practices of contact with poultry that give rise to the greatest H5N1 transmission risk potential. Of the 3,600 subjects included in this assessment, approximately 16.2% (n=583) had exposure risk scores above the 90th percentile and were largely male (72.3%) and had a median age of 29 (IQR: 21-42; range 6-69). These rural subjects have the greatest potential non-occupational risk for poultry-to-human transmission of HPAI.

As discussed in Chapter 5, this population group differs from the age and sex distribution of the total number of confirmed H5N1 human cases that occurred up to 30 December 2008, in which an excess of cases was observed in children and no differences observed between genders (Writing Committee of the Second World Health Organization Consultation on Clinical Aspects of Human Infection with Avian Influenza 2008). However the group with highest exposure in this study is more similar to the age/sex distribution of the confirmed Thai cases (n=25). The mean age of cases was 22 years and 64% of cases were male (WHO 2006-2009) indicating that similar poultry contact patterns may exist in Thailand.

Such socio-demographic differences in human cases of H5N1 may be because contact patterns with poultry differ between countries. However, it is also suggestive that the variation in H5N1 incidence by age may not be due to exposure alone and that there may be differences by age in intrinsic immunologic susceptibility to infection, pre-existing immunity against human influenza A virus and/or clinical presentation of disease. As my study is the first study to

evaluate individual-level behaviour of a large and randomly selected population, it would be useful to evaluate the poultry contact patterns of individuals living in rural areas of Vietnam, Thailand, Laos, China and other Asian and African countries to evaluate whether behaviour differs among these populations living in similarly close contact with poultry. No comparable studies have been conducted from which comparisons of regional contact patterns can be made, although contact with poultry is likely to be widespread.

This research found that poultry traders (i.e., poultry market sellers and middlemen) are in highly-concentrated contact with poultry and therefore have a greater potential poultry-to-human transmission risk as compared to rural Cambodians. All of the poultry traders included in my study reported practices that are considered to be high risk for effective transmission of H5N1 if a bird is infected with H5N1 since their daily activities regularly include contact with blood and bodily fluids through the practices of slaughtering, bleeding and handling internal organs of poultry without the use of personal protective equipment.

8.2 Limitations of the Research

There are several additional questions that I would have liked to include in the questionnaires to better understand the potential transmission risk of HPAI between flocks. For example, it would have been useful to obtain specific information on the grazing patterns of domestic duck flocks to evaluate if grazing patterns differ by region or among areas with reported outbreaks. It would also have been useful to address the frequency of use and extent of sharing of farm equipment. These data are needed to determine the risk of HPAI transmission within and between villages that share equipment and would be important to understand in order to design appropriate HPAI control strategies.

Secondly, I did not collect sufficient information to evaluate risk factors for within-flock mortality because of the nature in which the data on poultry mortality were collected. My study subjects had difficulty with the recall periods that were originally included in the questionnaires (e.g., *within the past two weeks, how many poultry died?*; *within the past two months, how many poultry died?*). Therefore, questions addressing poultry mortality captured the proportion of poultry that died over an 8-month period among the total number of poultry

owned during that same time period rather than the number of birds that died within each flock owned over a shorter, more specified time period.

Additionally, to better evaluate transmission risk of HPAI from poultry-to-humans, I would have liked to have obtained estimates of the extent and frequency of swimming/bathing/fishing in ponds by adults from all 6 provinces. I only collected information on swimming/bathing/fishing among residents in two provinces. Among these 800 adults, 97 reported regularly swimming/bathing/fishing in water sources where poultry have access. These practices were most often reported by 16-25 year old males. This information may provide insight on the age and sex distribution of the confirmed H5N1 cases that have occurred to date.

This research would have been enhanced if I had included a component in the study design to test for the presence of antibodies of H5N1 of the study participants. By testing my study subjects for anti-H5 antibodies, I may have been able to identify risk factors for infection. However in 2006, very few non-health care worker related studies had been conducted or resulted in finding any seropositive individuals (Bridges et al. 2002; Katz et al. 1999; Vong et al. 2006) and it was deemed not financially feasible to include this component in this study.

Similarly, my study of poultry movement would have been enhanced if I had included virologic and serologic testing of live poultry at the poultry markets included in my study (n=102). Given that NaVRI identified antibodies in approximately 8% of live bird samples from 13 markets in 2008, it is likely that I would have identified poultry with antibodies in some of the markets included in my study. These data could have been used to evaluate practices of the markets (e.g., how birds are kept, environmental conditions of the markets) to determine if any practices are associated with the presence of HPAI.

My semi-quantitative risk assessment had several limitations, largely because several epidemiologic data gaps remain. These data gaps include: a lack of understanding of the prevalence rates of H5N1 in poultry species in regions where H5N1 is endemic or recurrent; a lack of knowledge on all potential transmission routes from poultry-to-humans including by indirect means via the environment; a lack of understanding on the influence of genetic and/or immunological factors on transmission; and a lack of knowledge on the persistence of the virus

in poultry blood, bodily fluids, organs and tissues. As these data become available, the estimates of β may become more refined.

There are several limitations of my study of poultry traders. First, this study did not attempt to find and interview all poultry markets or all middlemen responsible for trading poultry in Cambodia. Thus, the results of the network analysis only represent poultry movement within my study areas, specifically within the 24 districts in 6 provinces included in the study. Although the connections into Phnom Penh may be fairly complete and represent regular trading into the capital, there are important hubs of poultry movement—particularly around Siem Reap—that would be important to uncover in order to provide more comprehensive active HPAI surveillance recommendations to NaVRI.

Secondly, although this study identified illegal cross-border poultry trading with Vietnam and Thailand, it did not capture the small-scale localized poultry trading that may be important in understanding the circulation of HPAI in the Mekong Delta Region. The extent of small scale, cross-border trading of poultry, which has been illegal since 2004, is not well understood but is occurring in many areas along Cambodia's permeable border with Vietnam, and in some cases involve district veterinarians in border areas (MAFF Unpublished Data). This movement needs to be fully examined before it can be controlled.

Phylogenetic analyses of circulating poultry and human H5N1 strains in Vietnam, Cambodia and Thailand since 2004 have indicated that H5N1 may have been introduced into Cambodia in early 2004 from Thailand and since then has been circulating back and forth across the Vietnamese/Cambodian border (IPC Unpublished Data). Phylogenetic analyses of H5N1 stains circulating in Vietnam from 2001 to 2007 suggest that there have been at least six introductions of H5N1 into Vietnam, possibly first introduced into Vietnam from cross-border trading activities with China (Wan et al. 2008). It is likely that the cross-border trading of small numbers of live backyard chickens and/or ducks may be a critical factor in HPAI circulation and persistence throughout the Mekong Delta Region.

8.3 Dissemination of Findings

The dissemination of study information throughout the various stages of the PhD—development, implementation, interim results and final results—was an important feature of my thesis work and therefore I regularly met with local partners to update them on the progress of the studies. For example, interim and final results of the cross-sectional study of rural Cambodians were presented to UNICEF in the form of an oral presentation and report at the end of the first phase (April 2007 (Van Kerkhove et al. 2007)) and second phase (March 2008 (Van Kerkhove et al. 2008)) of the study.

The results of this study have been used by UNICEF to improve their risk communication materials on avian influenza, specifically in school education. FAO Cambodia used the results on poultry mortality reporting to design a follow-up anthropological study to understand why subjects were reluctant to report poultry mortality to officials. IPC has used these results to design intervention studies in rural Cambodia to reduce the risk of transmission from poultry-to-humans by focusing on improvements in biosecurity on rural farms.

Detailed interim and final results of the cross-sectional study evaluating poultry movement were presented to local partners (IPC and NaVRI) in Cambodia in December 2007, October 2008 and March 2009 (planned) and provided as a report for NaVRI in early 2009. The results of this research have also been presented to members of Cambodia's avian influenza task force, which includes representatives from the WHO, FAO and the Ministry of Human Health in Cambodia.

8.4 Future Research

Despite numerous studies that have been published on H5N1 since 2003, there still remain important data gaps that must be filled to fully understand the epidemiology and pathogenicity of H5N1 in birds and humans. For example, risk factors for H5N1 human infection are still largely unknown and the persistence of H5N1 in different poultry species, within poultry populations and in the environment are not fully understood.

8.4.1 Persistence of H5N1 in Poultry Populations

There is a lack of knowledge of LPAI and HPAI virus persistence in poultry blood, bodily fluids, organs and tissues. To fully evaluate the transmission potential of H5N1 from poultry to humans, it would be useful to have a clear understanding of the pathogenicity of the circulating H5N1 strains in different species of vaccinated and unvaccinated birds, in assorted tissues and organs, and in the blood and bodily fluids at various days post infection. There have been numerous experimental studies that have evaluated the pathogenicity of H5N1 in different domestic and wild bird species, however these studies have largely been conducted on a small number of birds under controlled conditions (e.g., (Beato et al. 2007; Das et al. 2008; Hulse-Post et al. 2005; Spickler et al. 2008)). Since the pathogenicity of H5N1 in wild birds and domestic duck species has changed since 2003 (Pantin-Jackwood et al. 2007; Swayne 2007), it would be useful to have data collected from H5N1 outbreak investigations in domestic, commercial or wild bird populations (e.g., (Kwon et al. 2005)) as these data are essential for understanding the conditions that may be required for the effective transmission of H5N1 from poultry-to-humans.

There is also a lack of knowledge on the persistence of H5N1 strains and how transmission is sustained within poultry populations. There is likely considerable underreporting of HPAI in poultry in rural areas of many countries where backyard poultry is common (FAO Sector 4). It would therefore be useful to examine the poultry production systems of countries where H5N1 is present, for example by using the Sector 4 Sub-categories A through D that I described in Chapter 4. These sub-categories may be helpful in describing the poultry production systems where H5N1 outbreaks have been frequent (e.g., Vietnam, Thailand, Egypt) and less frequent (e.g., Laos, Malaysia, Benin) to determine if there is a relationship between husbandry practices and the frequency of H5N1 outbreaks or the risk of HPAI infection. While free-ranging duck flocks—which may be silent carriers of HPAI and have the potential to transmit the virus freely and undetected—are an important risk factor for the persistence of HPAI (Gilbert et al. 2006), semi-commercial farms may be at a higher risk for infection than backyard flocks (Graham et al. 2008; Sims 2007). However this could be the result of higher outbreak ascertainment in commercial poultry production settings (Graham et al. 2008). Therefore, understanding the poultry production system of countries with recurrent HPAI is vital for developing appropriate control programs.

The effects of control measures (e.g., vaccination, stamping out, restricting movement, improving bio-security measures), which have been implemented in many countries to control the spread of H5N1, are not well understood as there are few peer-reviewed and published reports that have evaluated such control programs. There is some information available from government and international organizational reports (MOH 2007; Otte et al. 2008) and presentations at conferences (Nguyen 2008; Siregar & Darminto 2008). However, it would be useful to obtain more specific information on which control strategies were used, the methods that were used to implement strategies, and results from post-vaccination surveillance and/or other evaluation programs.

The persistence of LPAI and HPAI strains in the environment under various climatic conditions—including those that resemble live bird markets, wet markets and poultry farms—is unclear. The often damp and dark conditions of wet markets are believed to be ideal for the persistence of H5N1 virus (Woo et al. 2006), and the conditions in the live birds selling areas and slaughtering areas could facilitate a viable environment for HPAI to survive and/or persist (Kung et al. 2007; Kung et al. 2003b; Nguyen et al. 2005; Wang et al. 2006; Webster 2004). There is some experimental evidence that suggests that influenza A viruses are detectable in water and wet faeces for up to 6 days at 37°C (Brown et al. 2007b) and that H5N1 can survive in carcasses for several days at room temperature (OIE 2008a; WHO 2006-2009). However further experimental studies are needed to evaluate the viability of H5N1 in various controlled climatic conditions with varying temperature, humidity, and UV light. These results would be useful to help understand the duration H5N1 is transmissible and detectable in various environments. Since my results found that birds spend less than 8 hours in market before they are slaughtered, it would be useful to evaluate the cost-benefit of including environmental sampling in HPAI surveillance programs conducted in markets, especially in resource limited countries.

8.4.2 Epidemiologic Data Gaps

Several important data gaps remain in the understanding of the epidemiology of H5N1 in humans. First, there remains considerable scope for underreporting of human and poultry H5N1 outbreaks and therefore data is still lacking on the risk factors for human H5N1 infection.

Since 2003, a small number of seroprevalence studies have evaluated the frequency of asymptomatic or subclinical infection and risk factors for H5N1 infection (Apisarnthanarak et al. 2005; Ortiz et al. 2007; Schultsz et al. 2005; Thanh Liem et al. 2005; Vong et al. 2009). However, only one of these studies has identified asymptomatic individuals with anti-H5N1 antibodies, indicating previous infection with H5N1. The interpretations from this study are limited because of the small numbers of cases (7) and matched controls (24) available for study.

It would be constructive to conduct a large-scale seroprevalence study of individuals who are in regular contact with poultry blood, tissues and bodily fluids. Given that anti-H5N1 antibodies were found in approximately 8% of live bird samples tested in 2008 (MAFF Unpublished Data), and that seroprevalence rates among birds at markets vary (Amonsin et al. 2008; Joannis et al. 2008; Wang et al. 2006) and could be higher, a human seroprevalence study should be conducted to evaluate the presence of HPAI (H5 and H7 subtypes) antibodies among poultry market sellers. This study should include poultry workers, especially individuals that are responsible for preparing poultry to be sold at the market (i.e., market sellers that slaughter, boil, defeather, handle internal organs, and cut meat) from multiple countries where H5N1 is recurrent. The inclusion of poultry market workers from several countries may allow for the evaluation of seroprevalence and/or risk factors by region. This study could evaluate the risk factors identified during my thesis work (and include risk factors evaluated in previous studies of poultry workers (Bridges et al. 2002; Ortiz et al. 2007)) and the extent and frequency of specific exposures to poultry that occur during the preparation of poultry for trading (e.g., slaughtering and bleeding poultry, boiling and defeathering poultry, removing and washing internal organs, etc) as well as other at risk practices involving direct and indirect contact with poultry in non-occupational settings. Careful attention to sample size should be considered when designing this study to adequately evaluate risk factors for infection since published studies have identified only a small percentage, if any, of individuals with anti-H5 antibodies (Bridges et al. 2002; Ortiz et al. 2007; Wang et al. 2006).

Second, the influence of genetic and/or immunological factors on transmission is poorly understood. Although there have been several suspected clusters of H5N1 infection (largely among blood relatives) where H5N1 may have been transmitted between humans, the clusters are difficult to interpret because all suspected family members may not have been tested for

H5N1. In an analysis of 11 suspected clusters of H5N1 among blood relatives in Indonesia, the authors suggest that there may have been limited human-to-human transmission in some clusters. However genetic variation in families could result in the occurrence of clusters because of a predisposition to infection (Kandun et al. 2008).

While no health care workers exposed to H5N1 patients in Vietnam or Thailand were infected from the care of these patients (Apisarnthanarak et al. 2005; Thanh Liem et al. 2005), a father may have been infected through contact during the care of his dying son infected with H5N1 at a hospital in China (Wang et al. 2008), and a mother and aunt may have become infected from similar contact with their dying daughter/niece in a hospital in Thailand (Ungchusak et al. 2005). In order to fully evaluate the occurrence of human-to-human transmission, a detailed exposure history needs to be collected from *all* suspected cases and their contacts. Bird and Farrar have developed a data collection form that could be used during all future human outbreak investigations, which includes not only information on contact with poultry and a suspect case, but includes questions regarding the timing of the contact (Bird & Farrar 2008). However this questionnaire covers only general exposure information (e.g., handling sick or dead poultry, handling faeces or fertiliser from sick or dead poultry, slaughtering poultry) and does not include any potential transmission via the environment (e.g., contaminated water). In order to build a database from which more robust analysis can be conducted, detailed exposure information should be systematically collected from all suspect cases.

Third, improved knowledge is needed on all potential routes of transmission of H5N1 from poultry-to-humans and the prevalence of such practices in human populations. I have evaluated what I believe are the main potential routes in which people can become infected with H5N1, but we currently lack sufficient data from the confirmed H5N1 cases around the world to fully evaluate other potential risk factors for infection such as the role of water and other environmental factors. Transmission could also include oral ingestion, conjunctival or intranasal inoculation from contaminated water while drinking, swimming or bathing or from faeces while caring for poultry (Vong et al. 2009) and may explain why more children than adults are infected. Furthermore, asymptomatic cases may occur because of low concentrations of viruses in the environment.

8.5 Conclusions

To fully understand and attempt to control a zoonotic disease like HPAI/H5N1, it is essential for animal and human health bodies to work effectively together to study HPAI in poultry and humans, especially at the animal/human interface. Collaboration between animal and human health sectors is essential to understand the risk of transmission between poultry and humans and to develop effective programs to control and prevent the spread of HPAI within poultry populations and onwards to humans. The exposure information that is currently collected from suspect H5N1 cases is too general to explain the current pattern or to predict future cases of H5N1 infection in human populations, and therefore need to be more systematic and standardized.

Outbreaks in human and poultry populations continue to occur. During the first two months of 2009, H5N1 was identified in humans in China and Egypt (WHO 2009a) and in poultry in Bangladesh, India, Nepal, Togo, China and Vietnam (OIE 2009b). H5N1 is often identified in human before poultry populations thus exposing the limitations of the current HPAI surveillance systems in many Asian countries where H5N1 is recurrent or endemic. Improvements in passive HPAI surveillance programs are urgently needed in these countries.

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Appendices

Appendix A: Published Manuscripts

Interaction Between Humans and Poultry, Rural Cambodia

Sowath Ly,* Maria D. Van Kerkhove,†
Davun Holl,‡ Yves Froehlich,§
and Sirenda Vong*

Because avian influenza H5N1 infection risks are associated with exposure to infected poultry, we conducted a knowledge, attitudes, and practices survey of poultry-handling behavior among villagers in rural Cambodia. Despite widespread knowledge of avian influenza and personal protection measures, most rural Cambodians still have a high level of at-risk poultry handling.

The circulation of the highly pathogenic H5N1 avian influenza (AI) strain throughout Asia since late 2003 (1), and more recently in Europe and Africa, has resulted in considerable concern for the potential of a new pandemic. In Cambodia, outbreaks of HPAI A/H5N1 infection were first reported in poultry in early 2004 (2). Since 2005, 6 human cases have occurred (100% fatal); the 2 most recent cases occurred in early 2006 (3,4).

Most Cambodians live in rural areas and raise animals for consumption (2), typically keeping poultry, swine, or cattle close to the home. Because H5N1 infection has been associated with exposure to infected poultry (5–10) and little is understood of the perceptions of rural farmers regarding AI (11), we conducted a knowledge, attitude, and practices survey of poultry handling in rural Cambodia to estimate the extent of interactions between humans and poultry, to understand practices in poultry handling among villagers, and to develop interventions designed to increase reports of poultry deaths and safe poultry handling.

The Study

We conducted a 2-stage household based cluster survey (12) with a goal of 500 participants: 20 persons ≥ 15 years of age in each of 25 villages from Prey Veng and Kampong Cham Provinces. The sampling frame of eligible villages within these provinces were those located in H5N1 high-risk communes, as defined by the Food and

Agriculture Organization of the United Nations training program for village animal health workers. The villages were selected with probability proportional to size. For the second stage, we randomly selected the first household within each village. Subsequently, households were selected by proximity until 20 eligible participants were enrolled in each cluster.

Verbal consent was obtained from all participants. All were interviewed by using a structured questionnaire designed to collect information on demographics, basic hygiene practices, quantity of poultry owned, poultry death reporting, practices when deaths occurred, knowledge and attitude of sick and dead poultry, and knowledge of AI.

Twenty-three villages were included in Kampong Cham (11) and Prey Veng (12) Provinces (Figure 1). Four hundred sixty respondents from 269 households completed the questionnaire. Most were women (60%), farmers (88%), and persons who had completed less than primary schooling (57%). The median number of household members was 5 (range 1–16), and 77% of all households included children < 15 years of age.

Many households owned chickens (97%) and ducks (39%) (Figure 2), although the size of most poultry flocks was small (Table). Almost all poultry were free ranging (100% of chicken flocks; 96% of duck flocks), and mixing of the poultry with pigs and other domestic animals was common. Respondents reported that they use poultry feces for manure (77%), touch sick/dead poultry with bare hands (75%), eat poultry that died from illness (45%), eat wild birds (33%), let children touch sick/dead poultry with bare hands (20%), and gather dead wild birds for consumption (8%).

During the previous 6 months, of the 260 households that owned poultry, 162 (62%) experienced poultry deaths;

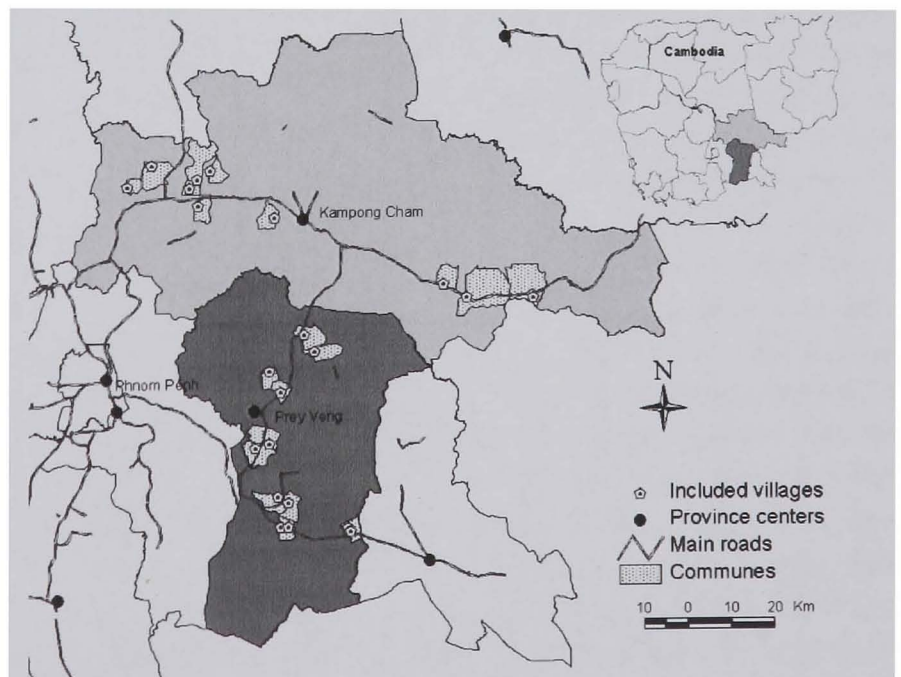


Figure 1. Distribution of selected communes in Kampong Cham and Prey Veng provinces, Cambodia, 2006.

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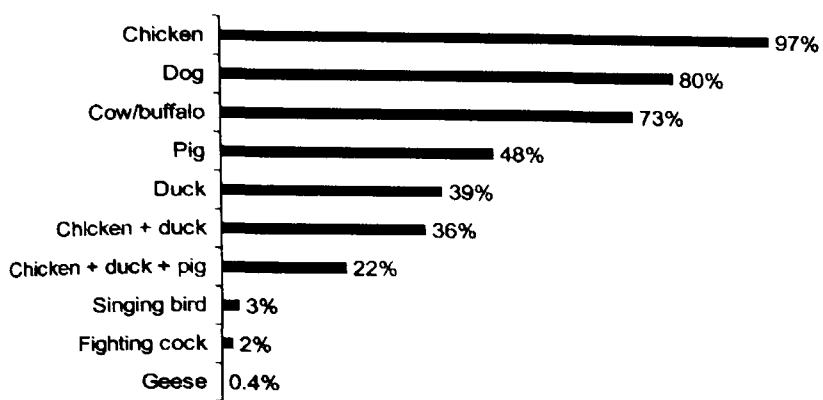


Figure 2. Proportions of animals raised in the household (n = 269), rural Cambodia.

however, only 18 (7%) reported these deaths to local authorities. Half of the respondents (n = 231) believed that it was important to report any poultry deaths because the death may be due to AI (61%) or because the poultry owners may receive management advice from the village veterinarians (39%). Among these 231 respondents, many did not report poultry deaths because they did not know how (41%), were in the habit of not reporting poultry deaths (31%), believed they would have a problem selling poultry if they reported deaths (18%), did not know the risks of AI (7%), or feared poultry culling (5%). Among those respondents who did not believe reporting deaths was important, the reasons provided included the following: “the number of poultry deaths were too few” (62%), “poultry are not as important as cattle” (18%), “no help would be provided from veterinary staff or authorities” (13%), or “because mortality was similar to previous years” (7%). Of respondents that experienced poultry deaths, 62% buried or burned dead poultry, 53% prepared them for food, 22% threw away the dead poultry, 3% used them to feed other animals, and 2% prepared them for sale or gave them to their neighbors.

Participants had learned about AI from television (81%) and radio (78%). Thirty-one percent of respondents

were able to describe AI symptoms in humans, and 72% believed that AI is a fatal disease among poultry that can be transmitted to humans. Most respondents believed it is unsafe to touch sick or dead poultry with bare hands (67%), eat wild birds (70%), let children touch sick or dead birds with bare hands (83%), and eat meat or eggs that are not fully cooked (86%). Sixty-one percent of respondents mentioned at least 1 of the recommended behavioral practices that protect against AI infection.

Conclusions

General media reports about AI through radio and television broadcasts appear to have been effective at reaching rural people. However, despite high awareness and widespread knowledge about AI and personal protection measures, most rural Cambodians still often practice at-risk poultry handling. Anecdotally, we also reported that family members of H5N1-infected patients, who knew about AI risks, still prepared dead or sick poultry for household consumption during massive die-offs, because they observed that neighbors with the same behavior did not become sick (Institute Pasteur in Cambodia, unpub. data). These findings provide evidence that high awareness does not necessarily lead to behavior change. Behavior change involves comprehensive and multidisciplinary intervention, which combines risk perception communication and feasible and practical recommendations, including economic considerations. We speculate that it is hardly feasible to sustain good poultry-handling practices if access to personal protective equipment is cost prohibitive, particularly when disease occurrence poultry die-offs are common. Further studies are needed to determine appropriate behavior change strategies in Cambodia.

We did find that many of the villagers were willing to report poultry deaths but did not know how. However, this finding should be interpreted in light of some limitations. We observed difficulties and frustrations among farmers whose flocks underwent culling after identification of H5N1 viruses in their flocks because compensation has not yet been approved by the government of Cambodia. In contrast, Thailand and Vietnam have introduced compensation along with the introduction of poultry vaccination in Vietnam and the reduction of backyard poultry ownership in Thailand in an effort to protect the commercial poultry industry. Thus, it is difficult to envision effective control strategies in Cambodia based exclusively on culling. Coincidentally, Vietnam has reported far fewer H5N1 outbreaks in poultry and humans since the introduction of the vaccination program, while Cambodia detected 4 outbreak sites in domestic poultry and 2 unrelated human cases in 2006. The real effect of a no-compensation policy on willingness to report poultry deaths needs to be assessed.

Flock characteristics	Chickens (n = 261)	Ducks (n = 97)
Median number per flock (range)	10 (1–110)	6 (1–800)
1–25	83%	93%
26–50	15%	3%
51–100	2%	0%
>100	0.4%	4%
Animal age when raising begins (months)	0 (0–15)	0 (0–12)
Type of raising		
Free ranging	100%	96%
Raising purpose (noncumulative)		
Household needs	73%	70%
Meat for sale	54%	42%
Eggs for sale	1%	16%

Not surprisingly, direct contact with poultry and poultry products was common among household members. Transmission of H5N1 from poultry to humans, even in circumstances in which human-poultry interactions are regular and intense has been limited; however, as the virus continues to circulate and evolve among poultry, bird-to-human transmission may increase. In this context, improvement in risky practices can only be achieved through relentless behavior change efforts. Because lack of knowledge does not appear to be a factor, intervention programs must include feasible options for resource-poor settings that have limited materials for personal protection (water, soap, rubber gloves, masks) and must offer farmers alternative methods to safely work with poultry on a daily basis.

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Frequency and patterns of contact with domestic poultry and potential risk of H5N1 transmission to humans living in rural Cambodia

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Background Since 2004, H5N1 outbreaks have been recurrent in domestic poultry and humans in Cambodia. To date, seven human cases (100% CFR) and 22 outbreaks in poultry have been confirmed. Household ownership of backyard poultry (FAO Sector 4 poultry production) in rural Cambodia is high. An understanding of the extent and frequency of poultry handling behaviors in these settings is necessary to assess the risk associated with different practices and to formulate sensible recommendations to mitigate this risk. We collected new data from six geographic regions to examine patterns of human contact with poultry among rural farmers in Cambodia and identify populations with the highest potential exposure to H5N1.

Methods and Findings A cross-sectional survey was undertaken in which 3,600 backyard poultry owners from 115 randomly selected villages in six provinces throughout Cambodia were interviewed. Using risk assessment methods, patterns of contact with poultry as surrogate measures of exposure to H5N1 were used to generate risk indices of potential H5N1 transmission to different populations in contact with poultry. Estimates of human exposure risk for each study participant ($n = 3600$) were obtained by multiplying each reported practice with a transmission risk-weighting factor and summing these over all practices reported by

each individual. Exposure risk estimates were then examined stratified by age and gender. Subjects reported high contact with domestic poultry (chickens and ducks) through the daily care and food preparation practices, however contact patterns varied by gender and age. Males between the ages of 26-40 reported practices of contact with poultry that give rise to the highest H5N1 transmission risk potential, followed closely by males between the ages of 16-25. Overall, males had a higher exposure risk potential than females across all age groups ($p < 0.001$).

Conclusions Our results demonstrate that most of the population in rural Cambodia is in frequent contact with domestic poultry. About half of the population in this study carried out on a regular basis at least one of the practices considered to be high risk for the effective transmission if the bird is infected. There was however substantial variation in the frequency of different practices and thus the potential risk of transmission of H5N1 from poultry to humans is not uniform across age and gender even amongst populations living in close proximity to poultry.

Keywords Animal-human interface, Cambodia, H5N1, risk analysis, semi-quantitative risk assessment, transmission risk.

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Background

Since late 2003, highly pathogenic avian influenza (HPAI/H5N1) has spread globally within wild and domestic bird populations and now appears endemic in many parts of Asia and Africa. Millions of people in South-East Asia and around the world live in close proximity to domestic poultry and although direct contact with infected birds is assumed to be the main source of

infection to humans,¹ neither the specific mode of effective transmission from animal to human nor the role of water or other environmental factors² is fully understood. Transmission of H5N1 from poultry to human is thought to most likely occur following direct contact with infected poultry organ tissue, blood, nasopharyngeal secretions or faeces under poor hygienic conditions; however, it could also include ingestion of contaminated water.³ The risk of transmission will be influenced by the nature and

frequency of contact with contaminated cells, tissue, blood or secretions in which the virus replicates.⁴⁻⁶ Most of the H5N1 laboratory-confirmed human cases to date have reported recent contact with infected poultry although the specific nature of the contact was not recorded.⁷ At present there are an excess of reported cases in children and young adults.⁸ However, in the absence of detailed exposure data it is not possible to ascertain whether these represent increased exposure, susceptibility to infection, susceptibility to severe disease or a combination of all three. To date three case-control studies have been conducted in Thailand, Vietnam and Cambodia^{2,9-10} to explore risk factors for infection. Exposure from the preparation of sick and dying poultry was noted as an important risk factor in one study¹⁰ but only 38% of the population risk of AI could be attributable to this exposure because of the relatively low prevalence of reporting of this practice. However, the power of these studies is limited because of their small sample size. In addition, there is a lack of reference data on how preparation of sick and dying poultry and other potential exposures differ within and between countries.

Within Cambodia, H5N1 outbreaks have been recurrent since 2004 in domestic poultry and humans. To date, seven human cases, all of which have been fatal,¹¹ and 22 outbreaks in poultry have been confirmed in villages mainly located in Southern Cambodia. Household ownership of backyard poultry (FAO Sector 4 poultry production) in

rural Cambodia is high.¹² An understanding of the extent and frequency of poultry handling behaviours in backyard poultry farming settings is necessary to assess the risk associated with different practices and formulate sensible recommendations to mitigate this risk. Here we present data collected from six geographical regions in Cambodia in which we explore patterns of human contact with poultry among rural farmers to identify populations with the highest H5N1 (or other subtypes of avian influenza) exposure potential.

Methods

Risk assessment framework

A conceptual pathway was developed within the risk assessment framework^{13,14} and is illustrated in Figure 1. It describes the steps to infection with H5N1 in humans from contact with poultry. The pathway includes the probability that an animal is infected with H5N1 (P), the probability that an individual comes in contact with an infected animal (C), and the probability of effective transmission of H5N1 from poultry to human in the absence of protective clothing (β).

Several important data gaps and uncertainties currently exist – namely the persistence of H5N1 in domestic/wild poultry populations and in the environment under different atmospheric conditions, virus survival in poultry species during food preparation practices, exposure

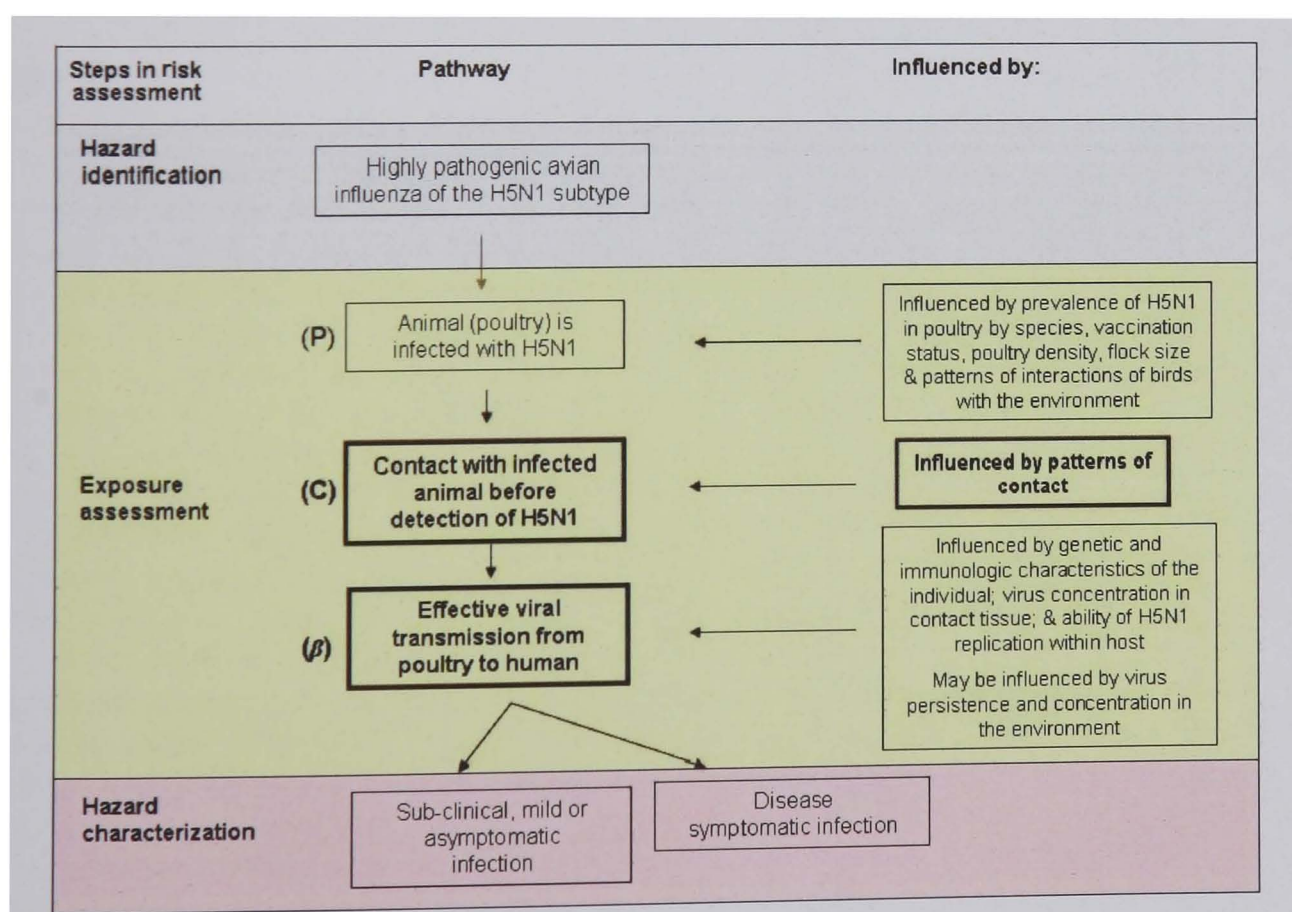


Figure 1. Conceptual pathway for transmission of H5N1 from poultry to humans via contact with poultry.

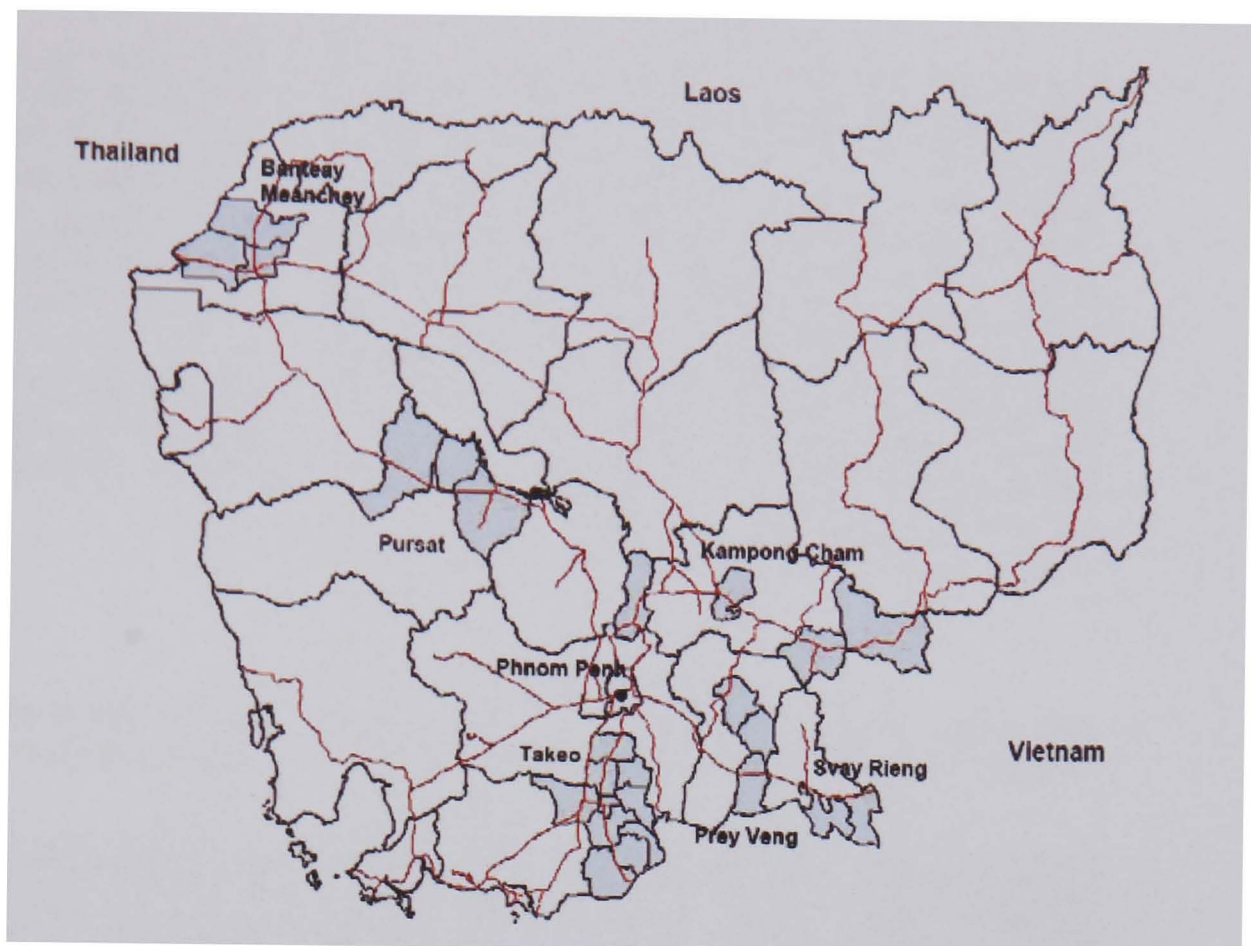


Figure 2. Study areas (districts shaded in grey, national roads indicated in red).

quantification of H5N1 from poultry and empirical data on risk factors for transmission from poultry to humans – making it difficult to perform a complete quantitative risk assessment.^{15,16} In this analysis, we contribute new field data to help with such an assessment focusing on the modules outlined in bold (patterns of contact that could result in effective transmission).

Data collection

A cross-sectional survey was carried out in six provinces using a two-stage clustered sampling method.¹⁷ Provinces and districts were identified for inclusion in the study from a preliminary assessment of high poultry ownership and human population density;¹⁸ potential cross-border trading activities, and wild bird mixing (Figure 2). H5N1 has not been suspected nor confirmed in poultry or humans in any of the 115 villages in the study areas; however, it has been confirmed in poultry and humans in one district in Kampong Cham and one district in Prey Veng Province.⁷ A random sample of 20 villages per province were selected using probability proportion to population size methodology (village population range 100 to >24 000).¹⁷ Subsequent households were then systematically sampled using a sampling interval having been chosen at random for each village until 30 people [10 male adults (>15 years old), 10 female adults (>15 years old) and 10 children (≤15 years old)] plus one village chief were interviewed. Individuals ≥1 year old, resident in village ≥6 months and medically fit to be interviewed directly or via an adult guardian were included. Ethical approval was granted from the Cambo-

dian Ministry of Health and London School of Hygiene and Tropical Medicine. Prior to sampling, field visits were conducted and meetings were held with provincial veterinarians and village chiefs. Sixteen Cambodian interviewers were trained to administer the questionnaires in Khmer. Informed written consent was obtained from all subjects or their guardians prior to interview.

Three separate standardized closed-ended questionnaires for the head of household, adult family members and children were administered to collect information on the types of direct and indirect contact with domestic and wild poultry. Heads of households were asked about poultry and other animal ownership (quantity of animals owned, husbandry practices, selling/trading practices) while all subjects, including all adults and children, were asked if they had direct contact with domestic poultry through food preparation (slaughter poultry, remove or clean internal organs, cut or wash meat) or other activities (e.g. collect dead domestic/wild poultry for food, eat wild birds, remove feathers from sick poultry, attend fighting cock events), cared for domestic poultry or fighting cocks (feed, clean animals or cages), and in the case of children, played with domestic and/or wild poultry. The nature of how Cambodians prepare poultry for consumption was evaluated by direct observation and informal questioning of adults living in rural Cambodia by the researchers (M.V.K., S.L.) in the field prior to piloting the questionnaires. The questionnaires for all subjects also asked if they had indirect contact with poultry – as a proxy measure of exposure – in the immediate environment around the home and vil-

lage via water sources (e.g. bathe/swim in ponds where poultry had access). Subjects were asked to recall practices within the previous 8 months, i.e. between the time of the interview and the Khmer New Year Holiday period (April 15). All responses to poultry contact questions were recorded as binary (yes/no) responses and frequencies of contact (when evaluated) were recorded as always, sometimes or never.

Questionnaires were checked daily and discrepancies checked with interviewers/observers prior to double entry into EpiData v3.1 (EpiData Association, Odense, Denmark).

Statistical methods

Prevalence of poultry handling behaviours

Poultry contact patterns were analysed by gender and age using chi-squared tests or Fisher's exact tests as appropriate.

ate. As a large number of food preparation variables were obtained, principal components analysis (PCA) was used to identify key practices that accounted for the variation observed across the population. Using PCA, a set of eigenvectors and eigenvalues were calculated for each of the factors (i.e. slaughter, boil, remove/wash internal organs, wash/cut meat) describing food preparation. Each principal component is a weighted combination of the original variables. Scree plots¹⁹ were used to retain those components contributing substantially to the overall sample variation. The newly created practice scores created from these principal components were subsequently analysed by gender and age group using *t*-tests or Wilcoxon rank-sum tests as appropriate.

Estimates of human exposure risk

Risk profiles were generated for each subject using their individual poultry handling contact patterns. The probabil-

Table 1. Prevalence of practice associated with poultry in rural Cambodian households, main sources of potential exposure and weighted transmission risk potential (β) ($n = 3600$)

Probability of effective viral transmission (β grouping)	Practice	Adult males	Adult females	Children	P-value	Adult males versus adult females	Adults versus children	Potential viral exposure
		($n = 1201$) >15 years old	($n = 1199$)	($n = 1200$) ≤15 years old				
High (β_1)	Remove internal organs (poultry)	733 (61.0)	588 (49.0)	156 (13.0)	<0.001	<0.001		O, B
	Blow into beak (FC)	19 (1.6)	1 (0.1)	6 (0.5)	<0.001		0.27	NS, B
	Kiss, suck, lick wounds (FC)	10 (0.8)	0 (0)	6 (0.5)	0.002		0.72	B
	Share water from the same bottle (FC)	21 (1.8)	4 (0.3)	21 (1.75)	0.001		0.07	NS, B
	Clean trachea (FC)	44 (3.7)	1 (0.1)	16 (1.3)	<0.001		0.235	NS, B
	Clean feathers (FC)	52 (4.3)	6 (0.5)	34 (2.8)	<0.001		0.46	B, F
	Wash internal organs (poultry)	745 (62.0)	775 (64.6)	249 (20.0)	0.185		<0.001	O, B
	Slaughter poultry	655 (54.5)	224 (18.7)	138 (11.5)	<0.001		<0.001	B, F
Moderate (β_2)	Touch/play with sick poultry or poultry that died from illness	597 (49.7)	485 (40.5)	90 (7.5)	<0.001		<0.001	B, F
	Use poultry faeces as manure	664 (55.3)	678 (56.6)	–	0.534		–	F
	Cut poultry meat	716 (59.6)	917 (76.5)	152 (12.7)	<0.001		<0.001	B
	Wash poultry meat	772 (64.3)	906 (75.6)	234 (19.5)	<0.001		<0.001	B
	Swim/bathe in water source where poultry have access*	56 (14.0)	41 (10.3)	196 (16.3)	0.113		0.01	F, NS
	Remove feathers from sick poultry*	76 (19.0)	101 (25.3)	102 (8.5)	0.04		0.001	NS, B, F
	Cleaning/sweeping poultry areas	843 (70.2)	903 (75.1)	442 (36.8)	0.005		<0.001	F
	Shopping at wet/live market for poultry	141 (11.7)	126 (10.5)	–	0.341		–	B, F
Low (β_3)	Boil poultry	673 (56.0)	898 (74.9)	228 (19.0)	<0.001		<0.001	B, F
	Living in a household with poultry (raised chickens or ducks within previous 8 months)	517 (86.7)**		1039 (86.6)	–		0.81	F

Values are expressed as n (%). FC, fighting cocks; B, blood, F, faeces; NS, nasopharyngeal secretions; O, organ tissue; –, not assessed.

*This practice was only evaluated in adults from two provinces ($n = 400$ adult males and 400 adult females).

**Evaluated from head of household questionnaire only ($n = 600$).

ity of effective viral transmission following a certain type of contact is assumed to be high, moderate or low as indicated in Table 1. A transmission risk weighting score (β) was applied to quantify the risk associated with high and moderate practices compared with low practices. Practices listed in group 1 are believed to have a higher potential transmission risk based on the nature of contact and potential H5N1 exposure than practices listed in 2 or 3 whereas practices listed in group 2 have a higher potential transmission risk than practices in group 3. In the analysis presented here, we used values $\beta_1 = 10$, $\beta_2 = 2$ and $\beta_3 = 1$. These values for β_1 , β_2 and β_3 are used in this analysis as an illustration of weighting exposures and are based on available data on the pathogenicity of H5N1 in poultry tissues.^{3,4,6,20-25} As more epidemiologic and virologic data about the persistence of H5N1 in poultry are collected, more precise estimates for these values may become available. Estimates of human exposure risk for each study participant ($n = 3600$) were then obtained by multiplying each reported practice with the transmission risk-weighting factor and summing these over all practices reported by each individual ($\sum\beta C$). The exposure risks were analysed by age and gender using *t*-tests or Wilcoxon rank-sum tests as appropriate. *P*-values of <0.05 were considered statistically significant. All statistical analyses were performed using Stata (v 9.2) (StataCorp, College Station, TX, USA).

Results

Poultry handling behaviours of adults and children

A total of 3600 household members [1200 adult (>15 years old) males, adult (>15) females and children (≤ 15)] were interviewed. The refusal rate was low ($<1\%$). The median age of adult and child subjects was 36 years (range: 16–87) and 9 years (range 1–15), respectively. The prevalence of poultry ownership is high in the study areas with 83.7% of households owning chickens, 35.7% owning ducks and

33.2% owning both chickens and ducks, although most poultry flocks are small [median chicken flock size (interquartile range IQR) = 14 (7–25); duck = 7 (3–15)].

Fighting cock ownership is low (3.8%), whereas ownership of pigs (55%), cattle/water buffalo (63.5%) and dogs (75.5%) is high. Mixing of domestic animals (53.8% of households owned pigs and poultry) is common. In rural areas of Cambodia, chickens and ducks are primarily raised for household consumption. Approximately 11% of adults reported shopping in wet/live markets for poultry. Few households reported selling domestic chickens [3.8% (23/600)] or ducks [0.5% (3/600)] outside their home village or to a market during the previous 8 months. Contact patterns with domestic poultry are provided in Table 1.

Food preparation practices

Preparing poultry for consumption consists of a series of steps including slaughtering the animal by breaking the neck or cutting the throat, bleeding, boiling, defeathering, removing and washing internal organs, and cutting and washing meat. Although family members as young as 2 years old reported that they had prepared poultry for consumption during the study periods, these practices were primarily the responsibility of family members 16–60 years old (Figure 3).

Both men and women were involved in each stage of preparation (Figure 2); however, overall, the proportion of adults involved in all practices related to food preparation was higher than children. Among adults ($n = 2400$) significantly more men than women slaughter poultry and remove internal organs whereas adult women more often boil poultry, cut meat and wash meat.

Among children, more males than females practice slaughtering (17.0% versus 5.8%, $P < 0.001$) and removing internal organs (15.7% versus 10.2%, $P = 0.005$), while more females than males are responsible for boiling poultry (22.3% versus 15.9%, $P = 0.005$) and cutting meat (15.7% versus 9.8%, $P = 0.002$).

Steps in poultry preparation

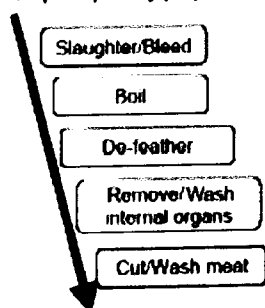


Figure 3. Food preparation practices by age group ($N = 3600$).

Practice	Adults ≥ 16 years old (% reporting)				Children < 16 years old (% reporting)			
	Total ($n = 2400$)	Male ($n = 1201$)	Female ($n = 1199$)	χ^2 (adult male v adult female)	p value	Total ($n = 1200$)	χ^2 (adult v child)	p value
Slaughter/bleed	36.6	54.5	18.7	332.3	<0.001	11.5	249.1	<0.001
Boil	65.5	56.0	74.9	94.4	<0.001	19.0	690.7	<0.001
Remove internal organs	56.0	61.0	49.0	34.9	<0.001	13.0	584.4	<0.001
Wash internal organs	63.3	62.0	64.6	1.7	0.185	20.0	601.7	<0.001
Cut meat	66.0	59.6	76.5	78.5	<0.001	12.7	581.3	<0.001
Wash meat	69.9	64.3	75.6	36.3	<0.001	19.5	816.6	<0.001

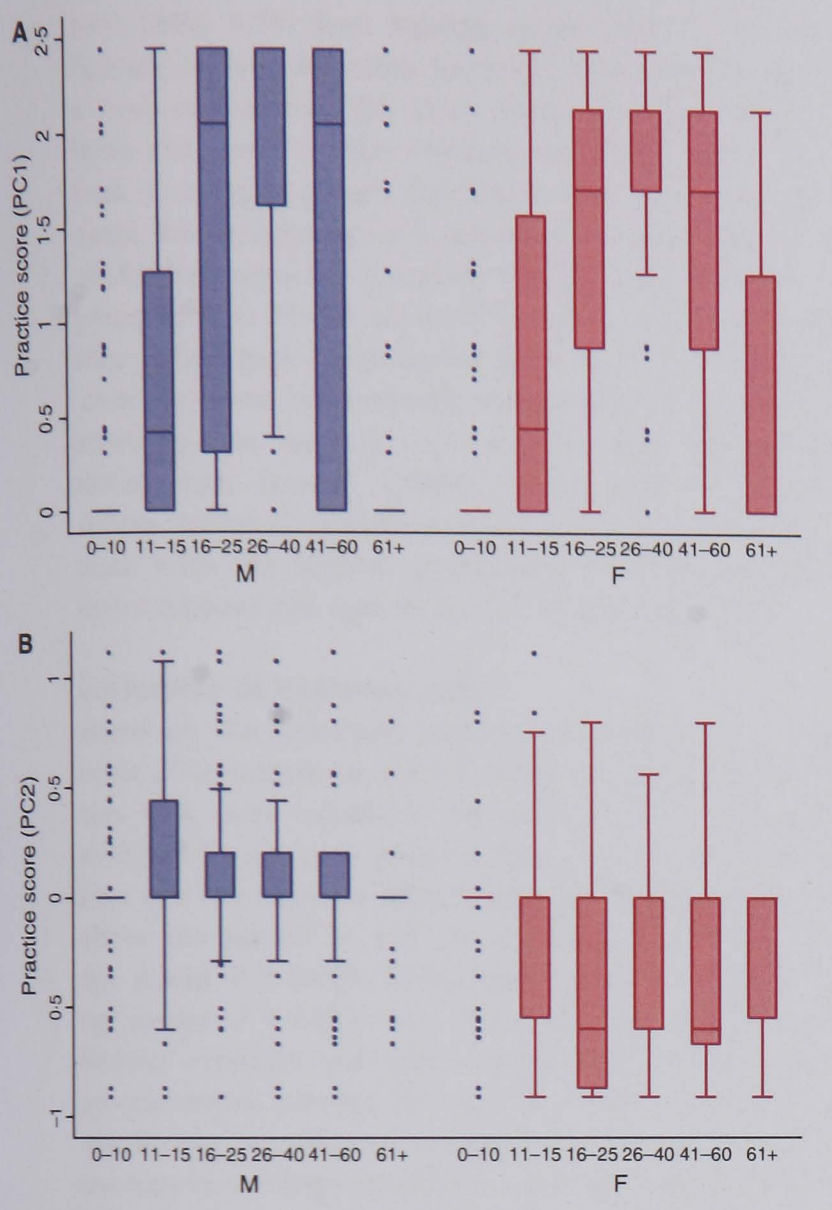


Figure 4. (A) Practice 1 – general food preparation by age and gender. (B) Practice 2 – slaughtering and removing internal organs by age and gender.

Principal component analysis of food preparation variables

The first principal component (practice 1), which accounts for approximately 71% of the total variation in practices between individuals in the survey, consisted of all six of the original food preparation variables (boil, slaughter, cut meat, wash meat, remove internal organs, wash organs) and hence can be interpreted as general food preparation. The second principal component (practice 2), which accounts for a further 13% of the variation, was dominated by the practices of slaughtering and removing internal organs.

The frequency of practice 1 (general food preparation, 71% variation) follows a similar age pattern in males and females with the highest scores between the ages of 16–25, 26–40 and 41–60 (Figure 4A). Subjects >60 years old had lower practice scores than children between the ages of 11 and 15 years.

Practice 2 (slaughtering and removing internal organs) shows greater differences by gender with this practice predominately undertaken by males (Figure 4B). There are sig-

nificant differences in practice 2 by gender among subjects with males reporting higher scores than females across all age groupings (two-sample *t*-test $P < 0.001$).

Other poultry contacts of adults and children

Regular contact with poultry for adult subjects ($n = 2400$) also includes using faeces for manure (56.6%; no variation by gender), touching sick or dead poultry with bare hands (49.7% in males versus 40.6% in females, $P < 0.001$), caring for fighting cocks (5.0% in males versus 1.4% in females, $P < 0.001$), and preparing wild birds for food (36.4% in males versus 19.3% in females, $P < 0.001$).

Among children ($n = 1200$) household responsibilities include feeding poultry (77.3%), gathering poultry and placing in designated areas or cages (43.5%), gathering/touching eggs (45.6%), cleaning poultry faeces (44.2% in males versus 37.4% in females, $P = 0.02$) and treating sick poultry with traditional medicines (18.5%).

Within the recall period, 35.9% of children reported that they had usually played with birds that were alive (42.5% male versus 29.0% female, $P < 0.001$), 2.7% reported playing with sick birds and 4.2% reported playing with dead birds (no gender difference). Thirty-two per cent of children removed feathers from sick/dead birds (no gender difference) and 16.3% of children bathed or swam in ponds (no gender difference) in which poultry had access; of those 37.8% reported doing this every day. Twelve per cent of adults ($n = 799$) reported swimming, bathing or fishing in ponds where poultry have access. Among all subjects who responded to this question ($n = 1999$), there are no gender differences in reported swimming/bathing in ponds; however, this reported activity was highest in children between the ages of 11 and 15 (16.5%) followed by children between the ages of 1 and 10 (16.2%) compared with adults.

A small number of children were involved in the care of fighting cocks (5.7%; $n = 68$; Table 1). Among children

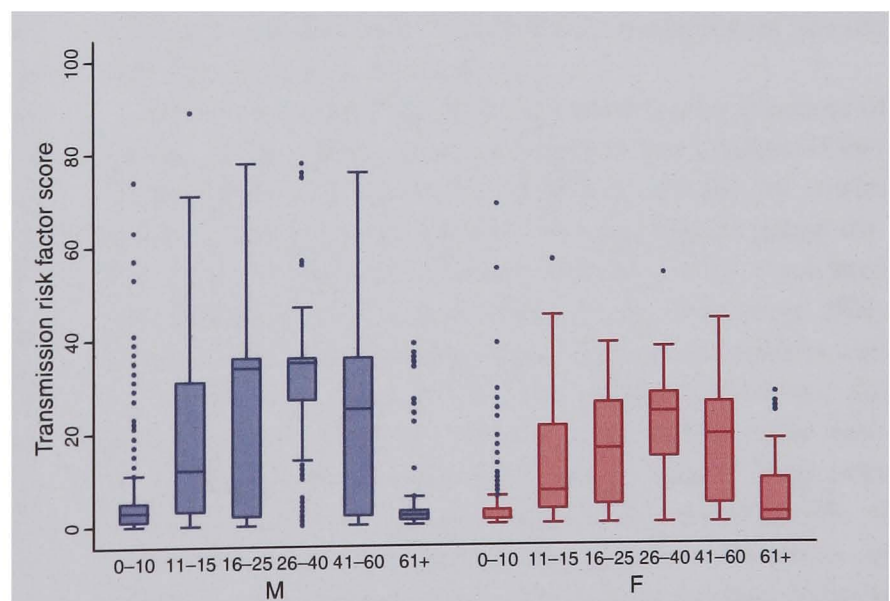


Figure 5. Exposure risk scores by age and gender.

($n = 1200$) 6.7% feed fighting cocks; 2.6% touch bloody fighting cocks; 2.8% clean feathers; 1.3% clean trachea with a swab or feather; 1.8% share water from the same bottle; 0.5% kiss, suck or lick wounds; and 0.5% blow into the beak of a fighting cock (the latter three are practices that occur during fighting cock matches). Twenty-eight per cent of children reported attending fighting cock matches compared with 11.3% of adults ($P < 0.001$) (Correction added after publication 20 November 2008: the words 'adults' and 'children' were inadvertently transposed). Among children, attendance at fighting cock matches was higher among males than females (35.0% versus 20.6%; $P < 0.001$). Adults reported attending matches on average once per week with the highest proportion of attendance among males between the ages of 16 and 25 years (31.7%).

Estimates of exposure risk

Based on the identified patterns of contact and assumptions of transmission risk (β ; Table 1), estimates of exposure risk were calculated for each subject and analysed stratified by age and gender (Figure 5). Overall, the exposure risk was higher among males than females for subjects above the age of 10 (11–15 age group, $P = 0.002$; 16–25 age group $P < 0.001$; 26–40 age group, $P < 0.001$; 41–60 age group, $P < 0.001$; 61+, $P < 0.001$). In both males and females exposure risk varies by age with the greatest risks among males between the ages of 26–40 and 16–25 (Figure 5). We also observed a high degree of variability in risk (as seen in the large confidence intervals). Of the 3600 subjects, there were 590 subjects with an exposure risk score above the 90th percentile of the sample. These subjects were predominately male (72.6%) with a median age of 30 (IQR range 21–42).

Discussion

Our results demonstrate that most of the population in rural Cambodia is in frequent contact with domestic poultry, with an estimated 52% of the population carrying out on a regular basis at least one of the practices that we considered of high risk of effective transmission if the bird is infected. We also found that the frequency of exposure to poultry was higher in our study population than that reported in the control subjects used in the Vietnamese¹⁰ and Thai⁹ case-control studies, suggesting that contact patterns in Cambodia may differ from those in these neighbouring countries. However, at present there are no other similar studies from these countries to enable a direct comparison to be made. Given the widespread exposure to poultry, it is perhaps surprising that only a small number of H5N1 cases have been reported in Cambodia (seven to date). Although there is considerable scope for under-reporting of human cases the small number of cases may

be due to several factors – the lower density of poultry per km² in Cambodia compared to Thailand and Vietnam,²⁶ the low probability of people dealing with an infected domestic bird (i.e. low H5N1 prevalence and/or a short duration of infectiousness), and a low probability of effective viral transmission.

Within Cambodia, the typical diet consists primarily of white rice and fish products; animal products compose less than 8% of the daily energy supply.²⁴ Eating poultry as a source of protein is usually reserved for special occasions, typically weddings and national holidays [e.g. Khmer New Year (April), Chinese New Year (January/February)] and food preparation of poultry therefore differs seasonally.

It is assumed that the probability of risk from preparing and consuming poultry is negligible if food preparation is conducted under strict hygienic conditions.¹⁶ The use of personal protective equipment (i.e. gloves, rubber boots, face masks, aprons) of the subjects in our study areas when in contact with poultry was negligible. Few individuals were in possession of these items in their homes with less than 5% of subjects reported wearing such items when handling poultry. Inactivation of H5N1 on the surface of poultry can occur when the animal is boiled, therefore if poultry are boiled before defeathering as is the case in Cambodia, the risk of exposure during defeathering is reduced. Furthermore, WHO guidelines state that cooking above temperatures of 70°C will inactivate H5N1 in meats and organs, therefore boiling before defeathering would also reduce exposure potential of individuals cutting/washing meat or internal organs.²⁷

Even though contact was widespread, there was substantial variation in the frequency of different practices, which although differing in magnitude according to practice provided evidence that the potential risk of transmission of H5N1 from poultry to humans is not uniform across age and gender even amongst populations living in close proximity to poultry. Public awareness campaigns and risk behaviour modification intervention programmes should therefore be targeted accordingly.

Males between the age of 26 and 40 reported practices of contact with poultry that give rise to the highest H5N1 transmission risk potential, followed closely by males between the age of 16 and 25. This population group differs from the age and sex distribution of the 357 confirmed H5N1 human cases that occurred up to 29 January 2008, in which an excess of cases were observed in children and no differences observed between genders; however, the group with the highest exposure in our study is more similar to the age/sex distribution of the confirmed Thai cases ($n = 25$). The mean age of cases was 22 years and 64% of cases were male.⁷ Such socio-demographic differences in human cases of H5N1 may be because contact patterns with poultry differ between countries; however, it is also

suggestive that the variation in H5N1 incidence by age may not be due to exposure alone and that there may be differences by age in intrinsic immunologic susceptibility to infection, pre-existing immunity against human influenza A virus and/or clinical presentation of disease.

This semi-quantitative risk assessment has several limitations and lacks the power of a formal quantitative risk assessment because of epidemiological data gaps and uncertainties of H5N1 pathogenesis in the host species. To improve future assessments a number of areas would need to be strengthened. First, data are urgently needed on the prevalence of H5N1 in poultry species in regions where H5N1 is recurrent or endemic in domestic poultry flocks. These data are likely to be influenced by the use of biosecurity measures on farms and in backyard farming settings. While H5N1 poultry outbreaks in countries are reported, because infection may remain asymptomatic in some host species (e.g. ducks), it is difficult to infer prevalence from poultry outbreak reports alone. Prevalence estimates in poultry will allow us to fully understand the probability that a farm or animal is infected with H5N1 (P, Figure 1).

Secondly, improved knowledge is needed on all the potential routes of transmission of H5N1 from poultry to humans and the prevalence of such practices in human populations. We have evaluated what we believe are the main potential routes in which people can become infected with H5N1; however, we currently lack sufficient data from the confirmed H5N1 cases around the world to fully evaluate other potential risk factors for infection such as the role water and other environmental factors play in transmission.²⁸ Transmission could also include oral ingestion, conjunctival or intranasal inoculation from contaminated water while drinking, swimming or bathing or from faeces while caring for poultry²⁹ and may explain why more children than adults are infected. Furthermore, asymptomatic cases may occur because of low concentrations of viruses in the environment.

Thirdly, an understanding of the influence of genetic and/or immunological factors on transmission is urgently needed since there has been limited yet inefficient human-to-human transmission.³⁰ Lastly, virus transmission potential should not be treated as equal across contact practices. Empirical data are needed on virus survival in poultry during food preparation practices, in poultry waste (i.e. poultry scrap, faeces), in soil and in water under different environmental conditions. In addition, data – either experimentally produced or collected during field investigations – are urgently needed on the persistence of H5N1 in poultry tissues. Specifically, which organ, tissue or secretion, if any, has the greatest potential for poultry-to-human transmission. One way of estimating this is to quantify the viral concentrations in various tissues under a variety of condi-

tions (e.g. days post-infection, whether or not the animal is exhibiting symptoms, by vaccination status, etc.).

Collaboration between human and animal health sectors is essential to understand the risk of transmission between domestic poultry and humans. Current exposure estimates are too general to explain the current pattern or to predict future cases of H5N1 infection in human populations. Rapid, systematic and standardized collection of detailed information on poultry contact patterns in suspected human outbreaks of H5N1 would improve our understanding of transmission from poultry to humans.

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Appendix B: Questionnaires for rural Cambodians:

Village Chief

Head of Household

Adult Subjects

Child Subjects

H5N1 survey among backyard poultry farmers

Questionnaire for Village Chief

កំរងសំណួរ សំរាប់ប្រធានភូមិ

Today Date: ___/___/___ Interviewer's name _____

Village: ភូមិ _____ Province ខេត្ត _____

Instructions to Interviewer: ការណែនាំសំរាប់អ្នកសួរ

កំរងសំណួរនេះប្រើសំរាប់សួរប្រធានភូមិ... សូមអានគ្រប់សំណួរទាំងអស់ និង ធ្វើតាមការណែនាំក្នុងការរំលងសំណួរ ទៅកាន់ផ្នែកផ្សេងៗ ទៃកំរងសំណួរ... សូមគូសចំណែកលើរាល់សំណួរដែលបានសួរ និង សូមកុំរំលងសំណួរ បើមិនមានកំណត់សំគាល់ថាអោយរំលង។

1 Family Name: ឈ្មោះគ្រួសារ _____ 2 First Name: ឈ្មោះខ្លួន _____
3 Sex ភេទ 1 [] Male ប្រុស 2 [] Female ស្រី
4 Age អាយុ [] [] [] years old ឆ្នាំ
5 Address Village ភូមិ _____ 6 Occupation មុខរបរ _____
Commune ឃុំ _____ 7 Education level reached : កំរិតការសិក្សា
District ស្រុក _____ 0 [] Never attended school មិនដែលរៀនសោះ 3 [] High school វិទ្យាល័យ
Province ខេត្ត _____ 1 [] Primary បឋមសិក្សា 4 [] Higher ឧត្តមសិក្សា
2 [] Secondary មធ្យមសិក្សា 5 [] Pagoda រៀននៅវត្ត

8 In what country were you born? តើអ្នកកើតនៅទីណា?
1 [] Cambodia កម្ពុជា 2 [] Vietnam វៀតណាម 3 [] Thailand ថៃ 9 [] Other (specify) ផ្សេងបញ្ជាក់ _____
9 What is the primary religion of your village? តើប្រជាជនក្នុងភូមិរបស់អ្នក ភាគច្រើនកាន់សាសនាអ្វី?
1 [] Buddhist ព្រះពុទ្ធ 2 [] Muslim ឥស្លាម 3 [] Catholic សាសនាគ្រីស 9 [] Other (specify) ផ្សេងបញ្ជាក់ _____
10 How many people live in your village? តើភូមិរបស់អ្នកមានប្រជាជនប៉ុន្មាននាក់? [] [] [] [] [] people Year [] [] [] [] []
11 How many households are in your village? តើភូមិរបស់អ្នកមានផ្ទះប៉ុន្មានខ្នង? [] [] [] [] [] households Year [] [] [] [] []
12 When was this data collected? តើភ្នំលេខខាងលើ អ្នកស្រង់នៅពេលណា? _____

13 How many middlemen bought poultry from you village during the last month? [] 99 [] Don't know If 0 or DK, skip to 17
ក្នុងរយៈពេល ១ខែ ចុងក្រោយនេះ តើ មានអ្នកដើរទិញមាន់ទា តាមផ្ទះ ប៉ុន្មាននាក់ មកក្នុងភូមិរបស់អ្នក?
14 How often do they visit your village to purchase chickens and ducks? តើពួកគេចូលមកទិញមាន់ទាក្នុងភូមិរបស់អ្នក ប៉ុន្មានដង ក្នុង១អាទិត្យ?
[a] For chickens មាន់ [] [] [] [] times a week 99 [] Don't know
[b] For ducks ទា [] [] [] [] times a week 99 [] Don't know
15 Where do they take the poultry? តើពួកគេ យកមាន់ទា ដែលប្រមូលទិញបាន ទៅលក់បន្តនៅឯណាខ្លះ (ឈ្មោះផ្សារ, ភូមិ, ឃុំ, ស្រុក) 99 [] Don't know
[a] _____ [b] _____ [c] _____
16 Can you identify them by name and/or phone number? តើអ្នកស្គាល់ ឈ្មោះ ប្រភព ឬ លេខទូរស័ព្ទ របស់ពួកគេ ខ្លះទេ? 1 [] Yes, Continue 0 [] No, Go to Q#17
What are their names? From which villages? Do you know how to contact them?
ពួកគេឈ្មោះអ្វីខ្លះ? ពួកគេមកពីភូមិណាខ្លះ? លេខទូរស័ព្ទរបស់ពួកគេ?
[a] _____ [b] Village ភូមិ _____ [c] Phone _____
[d] _____ [e] Village ភូមិ _____ [f] Phone _____
[g] _____ [h] Village ភូមិ _____ [i] Phone _____
[j] _____ [k] Village ភូមិ _____ [l] Phone _____
[m] _____ [n] Village ភូមិ _____ [o] Phone _____
17 Would you be willing and able to find out this information? 1 [] Yes 0 [] No

18 How far is the nearest public health center (km)? តើ មណ្ឌលសុខភាព ដែលជិតជាងគេមានចំងាយប៉ុន្មានគីឡូម៉ែត្រ ពីទីនេះ? [] [] [] km
18a What is the name of this public health center? តើមណ្ឌលសុខភាពនោះឈ្មោះអ្វី? _____
18b What are the roads leading to the nearest public health center made of? តើផ្លូវទៅមណ្ឌលសុខភាពដែលជិតជាងគេនោះ កសាងអំពីអ្វី?
1 [] Dirt ផ្លូវដី 2 [] Paved ផ្លូវប្រាសបេតុង ឬ កៅស៊ូ 3 [] River ផ្លូវទឹក 9 [] Others ផ្សេងបញ្ជាក់ _____
19 How far is the nearest hospital (km)? តើ មន្ទីរពេទ្យដែលជិតជាងគេប៉ុន្មានគីឡូម៉ែត្រ ពីទីនេះ? [] [] [] km
19a What is the name of this hospital? តើមន្ទីរពេទ្យនោះឈ្មោះអ្វី? _____

Thank you for answering my questions អរគុណដែលបានឆ្លើយសំណួរ របស់យើងខ្ញុំ

H5N1 KAP survey among backyard poultry farmers
Questionnaire for Head of Household

កំរងសំណួរសំរាប់មេគ្រួសារ

Today Date: ____/____/____

Interviewer's name: _____

GPS | | | | | | | | | | | | | | | | | | | | | |

Province Village Family Subject number
ID Code | | | | | | | | | | | | | | | | | | | | | |

(PV=Prey Veng; KC = Kampong Cham; Exp. KC010105)

Instructions to Interviewer:

ការណែនាំសំរាប់អ្នកសួរ

កំរងសំណួរនេះប្រើសំរាប់សួរអ្នកដែលជាមេគ្រួសារ ។ មេគ្រួសារអាចជាបុរស ឬ ស្ត្រី ។ សូមអានគ្រប់សំណួរទាំងអស់ និង ធ្វើតាមការណែនាំក្នុងការលេខសំណួរ ទៅកាន់ផ្នែកផ្សេងៗ នៃកំរងសំណួរ ។
សូមត្រួតពិនិត្យលើលិខិតសំណួរដែលបានសួរ និង សូមកុំរំលងសំណួរ បើមិនមានកំណត់សំគាល់ថាអោយរំលង ។

សូមសរសេរលេខកូដ នៅផ្នែកខាងលើ ខាងស្តាំ ទំព័រនីមួយៗរបស់កំរងសំណួរ ។ ការសរសេរលេខកូដនេះមានសារៈសំខាន់ណាស់ ព្រោះថាទំព័រមុខរបស់កំរងសំណួរ នឹងត្រូវផ្តាច់ចេញ ហើយទុកដោយឡែក
មុននឹងធ្វើការបញ្ជូនទិន្នន័យ ។

ប្រសិនបើអ្នកមានចំលងផ្សេងៗទាក់ទងនឹងកំរងសំណួរ សូមទូរស័ព្ទទាក់ទងទៅ ឈ្មោះ ម៉ារ៉េ ដែលមានលេខ ០១២ ៨០២ ៤៧៥ ឬ ទៅ ឈ្មោះ សុវត្ថិ ដែលមានលេខ ០១២ ៣២២ ០៣១ ។
ពេលដែលបំពេញកំរងសំណួរចប់សព្វគ្រប់ សូមទុករក្សាទុកកំរងសំណួរជាមួយគ្នាដោយចាត់ទៅតាមផ្ទះនីមួយៗ រហូតដល់ចប់ថ្ងៃនីមួយៗ ។ បន្ទាប់ពីនោះទើប ទំព័រមុខនឹងត្រូវផ្តាច់ចេញពីកំរងសំណួរ ។

អ្នកសួរអានទៅអ្នកឆ្លើយ:

សូមអរគុណដែលបានយល់ព្រមចូលរួមក្នុងការសិក្សារបស់យើងខ្ញុំ ។ ជាដំបូងខ្ញុំសូមសួរសំណួរទាក់ទងនឹងខ្លួនអ្នកផ្ទាល់ និង គ្រួសាររបស់អ្នក ។ សូមមេត្តាជួយឆ្លើយឱ្យបានគ្រប់សំណួរតាមដែល អាចធ្វើទៅបាន ។
គ្រប់ចំលើយរបស់អ្នកទាំងអស់ នឹងត្រូវទុកជាការសម្ងាត់ ។ បើអ្នកមានចម្ងល់ ឬ មិនយល់សំណួរដែលខ្ញុំបានសួរ សូមប្រាប់ខ្ញុំ ខ្ញុំនឹងសួរជាថ្មី ។

ដូចនេះ ខ្ញុំសូមចាប់ផ្តើមសួរ:

Demographic information of respondent ព័ត៌មានផ្ទាល់ខ្លួន របស់អ្នកឆ្លើយ

Respondent: _____ (Record only surname)

1 Sex ភេទ 1[] Male ប្រុស 2[] Female ស្ត្រី

2 Age អាយុ _____ years old ឆ្នាំ

3 Address អាសយដ្ឋាន [a] Village ភូមិ _____ [c] Distict ស្រុក _____
[b] Commune ឃុំ _____ [d] Province ខេត្ត _____

4 Occupation មុខរបរ _____

5 Education level reached: កម្រិតសិក្សា
0[] Never attended school មិនដែលបានចូលរៀន 1[] Primary បឋមសិក្សា 2[] Secondary អនុវិទ្យាល័យ 3[] High school វិទ្យាល័យ 4[] Higher ឧត្តមសិក្សា 5[] Pagoda រៀននៅវត្ត

6 Can you read "I live in Cambodia"? សូមអ្នកអានឃ្លានេះ ខ្ញុំរស់នៅក្នុងប្រទេសកម្ពុជា 1[] Yes 0[] No

7 Can you write "Cambodia has many palm trees"? សូមអ្នកសរសេរឃ្លានេះ ប្រទេសកម្ពុជាមានដើមឆ្នោតច្រើន 1[] Yes 0[] No

8 Where were you born? 1[] Cambodia កម្ពុជា 2[] Vietman វៀតណាម 3[] Thailand ថៃ 9[] Other (specify) _____ ផ្សេងទៀត, បញ្ជាក់

9 What is your religion? 1[] Buddhist សាសនាព្រះពុទ្ធ 2[] Muslim សាសនាអ៊ីស្លាម 3[] Catholic សាសនាគ្រីស្ទ 9[] Other (specify) _____ ផ្សេងទៀត, បញ្ជាក់

សូមអាន ខ្ញុំរស់នៅក្នុងប្រទេសកម្ពុជា

សូមសរសេរ

SECTION A QUESTIONS TO HEAD OF FAMILY ONLY

សំណួរសំរាប់ស្រូវមេគ្រួសារ

How many people in total live in the house? តើសព្វថ្ងៃមានមនុស្សទាំងអស់ប៉ុន្មាននាក់រស់នៅក្នុងផ្ទះនេះ?

How many children 15 years old and younger live in the house? តើសព្វថ្ងៃ មានក្មេងអាយុ ចាប់ពី១៥ឆ្នាំ ចុះក្រោម ប៉ុន្មាននាក់រស់នៅក្នុងផ្ទះនេះ?

What is your house made of? តើផ្ទះរបស់អ្នកសាងសង់អំពីអ្វី?
 1 Wooden house ផ្ទះឈើ
 2 Cement house ផ្ទះថ្ម
 3 Straw/wood ផ្ទះស្លឹក
 4 wood and cement ឈើផង ថ្មផង
 9 Other (specify) _____ ផ្សេងទៀត, បញ្ជាក់

Is the house built on piles high up above the ground? តើផ្ទះរបស់អ្នកសាងសង់ មានជើងសសរ ខ្ពស់ជុំតពីដី ដែរឬទេ? 1 Yes 0 No

In your house, do you have (check all that apply)? ក្នុងផ្ទះរបស់អ្នក, តើមានអ្នកមានឧបករណ៍ដូចបានរៀបរាប់នេះដែរឬទេ?

[a] Bicycle កង់	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	[e] Telephone ទូរស័ព្ទ	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No
[b] Motorcycle ម៉ូតូ	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	[f] Television ទូរទស្សន៍	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No
[c] Car ឡាន	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	[g] Radio វិទ្យុ	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No
[d] Cart រទេះ	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No			

Is there an open water well in the houseyard? (observation) តើ នៅផ្ទះរបស់អ្នក មានអណ្តូងយោង ឬទេ? 1 Yes 0 No

Is there a water pump in the houseyard? (observation) តើ នៅផ្ទះរបស់អ្នក មានអណ្តូងស្នប់ ឬទេ? 1 Yes 0 No

Is there a pond in the houseyard? (observation) តើ នៅផ្ទះរបស់អ្នក មាន ត្រពាំង ឬ ស្រះទឹក ដែរឬទេ? 1 Yes 0 No

Is pond water used in your household for bathing? តើ អ្នកដងទឹកត្រពាំង / ស្រះ នោះ មកប្រើសំរាប់មុជ នៅក្នុងផ្ទះរបស់អ្នក ដែរឬទេ? 1 Yes 0 No

Is pond water used in your household for washing clothes? តើ អ្នកដងទឹកត្រពាំង / ស្រះ នោះ មកប្រើសំរាប់បោកគក់ខោអាវ នៅក្នុងផ្ទះរបស់អ្នក ដែរឬទេ? 1 Yes 0 No

Is pond water used in your household for cooking? តើ អ្នកដងទឹកត្រពាំង / ស្រះ នោះ មកប្រើសំរាប់ដាំស្ល នៅក្នុងផ្ទះរបស់អ្នក ដែរឬទេ? 1 Yes 0 No

Is pond water used for your animals (drinking, bathing, playing)? តើ អ្នកប្រើទឹកត្រពាំង / ស្រះ នោះ សំរាប់សត្វរបស់អ្នក ដែរឬទេ (ឧ. លាងគោក្របី, សត្វផឹក, ចុះលេង)? 1 Yes 0 No

Do ducks or geese have access to your pond (ducks from neighbourhood are applicable)? តើ មានសត្វទា ឬ ក្លាន ចុះលេង ក្នុងទឹកត្រពាំង / ស្រះ របស់អ្នក ដែរឬទេ? 1 Yes 0 No

Does your pond providd your family with food e.g. animals (fish, frog, shrimp, ...) or vegetables? តើ ត្រពាំង / ស្រះ មាន ត្រី ឬ សត្វ (កង្កែប, កំពឹស ...) ឬ បន្លែផ្សេងៗ សំរាប់គ្រួសាររបស់អ្នក យកមកធ្វើម្ហូប ដែរឬទេ? 1 Yes 0 No

Do you raise fish in your pond (natural fish is not applicable)? តើ អ្នកដាក់ចិញ្ចឹមត្រី នៅក្នុងត្រពាំង / ស្រះ នោះ ដែរឬទេ? 1 Yes 0 No

If NO, Go to A6

Now going to ask you some questions about your poultry flocks. ខ្ញុំសូមសួរបន្តទាក់ទងនឹងម្ចាស់ទាដែលអ្នកចិញ្ចឹម

What animals have you raised since the Khmer New Year? ចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក តើអ្នក មានចិញ្ចឹមសត្វអ្វីខ្លះ, ចំនួនប៉ុន្មាន?

- | | | |
|---|---|--|
| [i] Chickens មាន់ # <input type="text"/> | [e] Singing birds លលក, សេក, សារិកា ។ល។ # <input type="text"/> | [i] Pigs ជ្រូក # <input type="text"/> |
| [j] Ducks ទា # <input type="text"/> | [f] Pigeon ក្រាប # <input type="text"/> | [j] Dogs ឆ្កែ # <input type="text"/> |
| [k] Geese ក្លាន # <input type="text"/> | [g] Kruoch ក្រូច # <input type="text"/> | [k] Vaccinated dogs against rabies ឆ្កែដែលបានចាក់វ៉ាក់សាំងការពារជំងឺឆ្កែឆ្លុត # <input type="text"/> |
| [m] Other animal # <input type="text"/> Specify _____ | [h] Fighting cocks មាន់ជល់ # <input type="text"/> | [l] Cattle / buffalo គោ ឬ ក្របី # <input type="text"/> |
| [n] Other animal # <input type="text"/> Specify _____ | | |

skip if no chickens or ducks

Skip if respondent does not own ducks

Questions related to poultry raising	ការចិញ្ចឹម មាន់ និង ទា	Chickens មាន់	Ducks ទា
A7 What was their age at purchase (ave flock age in months)? តើនៅពេលដាក់ចិញ្ចឹមដំបូង សត្វមានអាយុប៉ុន្មាន?		a _____ weeks 99 <input type="checkbox"/> DK សប្តាហ៍ 0 <input type="checkbox"/> <1 week	a _____ weeks 99 <input type="checkbox"/> DK សប្តាហ៍ 0 <input type="checkbox"/> <1 week
A8 Where were they purchased? តើអ្នកទិញមកពីទីកន្លែងណាខ្លះ? (ចំណើយអាចមានច្រើន)	In your village ក្នុងភូមិដែលខ្ញុំរស់នៅ In your district ក្នុងស្រុកដែលខ្ញុំរស់នៅ (ភូមិផ្សេង) In your province ក្នុងខេត្តដែលខ្ញុំរស់នៅ (ស្រុកផ្សេង) In other province ទិញមកពីខេត្តផ្សេង In other country ទិញមកពីប្រទេសផ្សេង Born here from the last generation កើតនៅក្នុងផ្ទះរបស់ខ្ញុំតែម្តង Buy from market for raising ទិញពីផ្សារមកចិញ្ចឹម	b <input type="checkbox"/> c <input type="checkbox"/> d <input type="checkbox"/> e <input type="checkbox"/> Province: ខេត្ត _____ f <input type="checkbox"/> Country: ប្រទេស _____ g <input type="checkbox"/> h <input type="checkbox"/>	b <input type="checkbox"/> c <input type="checkbox"/> d <input type="checkbox"/> e <input type="checkbox"/> Province: ខេត្ត _____ f <input type="checkbox"/> Country: ប្រទេស _____ g <input type="checkbox"/> h <input type="checkbox"/>
A9 Type of raise : របៀបចិញ្ចឹមមាន់ទា (ចំណើយអាចមានច្រើន)	Birds always kept in closed building ចិញ្ចឹមដាក់ក្នុងទ្រុងបិទជិតជានិច្ច Birds raised in fenced park ចិញ្ចឹមដាក់ក្នុងទីធ្លាបិទជិត Birds free ranging in farmyard អោយដើររកស៊ីដោយសេរី ខាងក្នុង បរិវេណផ្ទះ Birds free ranging in and outside farmyard អោយដើររកស៊ីដោយសេរី ខាងក្នុង និង ខាងក្រៅ បរិវេណផ្ទះ Birds allowed to go to rice fields or lake កៀងដើររកស៊ីតាមវាលស្រែ ឬ តាមបឹង	i <input type="checkbox"/> j <input type="checkbox"/> k <input type="checkbox"/> l <input type="checkbox"/> m <input type="checkbox"/>	i <input type="checkbox"/> j <input type="checkbox"/> k <input type="checkbox"/> l <input type="checkbox"/> m <input type="checkbox"/>
A10 What was the lifespan of your last flock of poultry? ជាទូទៅ តើអ្នកចិញ្ចឹមមាន់ទា រយៈពេលប៉ុន្មានខែ ទើបអាចលក់ ឬ ហូបបាន?		n _____ months ខែ	n _____ months ខែ

1 Does your house have a fence around it? តើផ្ទះរបស់អ្នកមានរបងព័ទ្ធជុំវិញជិតដែរឬទេ?	1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No	If NO, Go to Q#A13
2 Do the fences keep your poultry inside your property? តើរបងព័ទ្ធជុំវិញអាចយ៉ាងជិតមិនឱ្យសត្វចេញចូលបានដែរឬទេ?	1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No	

Poultry selling practices ការលក់ដូរបសុបក្សី

Now going to ask you some questions about your poultry selling practices. ខ្ញុំសូមសួរបន្តទាក់ទងនឹងការលក់ដូរមាន់ទារបស់អ្នក

3 Do you sell chickens or ducks? តើអ្នកលក់ មាន់ ឬ ទា ដែរឬទេ?	1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No	If NO, skip to A32
4 [a] Since the Khmer New Year, have you sold CHICKENS from your home? គិតចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក នៅនឹងផ្ទះ តើអ្នកលក់ មាន់ ដែរឬទេ ? [b] How many times? តើអ្នកលក់ មាន់ បានប៉ុន្មានដងហើយ គិតចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក? _____ times since Khmer New Year ចំនួនលើក	1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No	If NO, skip to A19
[c] Do you sell chickens alive, prepared or both? តើលក់បែបណា? រស់, ធ្វើហើយ ឬ ទាំង២យ៉ាង?	1 <input type="checkbox"/> Only live 2 <input type="checkbox"/> Only prepared 3 <input type="checkbox"/> Both លក់តែរស់ លក់តែធ្វើហើយ ទាំង២យ៉ាង	

A15 To whom do you sell?
 តើអ្នកលក់ទៅឱ្យអ្នកណាខ្លះ?
 a People in your villager
 អ្នករស់នៅក្នុងភូមិជាមួយគ្នា
 b Middlemen from outside village
 ឈ្មួញមកពីខាងក្រៅភូមិ
 c Middlemen from Vietnam
 ឈ្មួញមកពី វៀតណាម
 d Middlemen from Thailand
 ឈ្មួញមកពី ថៃ
 e Other (specify)
 ផ្សេងទៀត _____

If used a middleman: បើអ្នកលក់ទៅឱ្យ ឈ្មួញដែលដើរទិញតាមផ្ទះ

A16 How many times since the Khmer New Year did you use a middleman to sell poultry [CHICKENS]?
 តិចចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក តើអ្នកលក់ មាន របស់អ្នក ទៅឱ្យឈ្មួញដែលដើរទិញ បានប៉ុន្មានដង?
 _____ times **If 0, skip to A19**

A17 How many middlemen did you sell to since Khmer New Year?
 តិចចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក តើ អ្នកលក់ មាន របស់អ្នក ទៅឱ្យ ឈ្មួញប៉ុន្មាននាក់?
 _____ middlemen 99 Don't know

A18 What is the name of the middlemen that you use?
 តើឈ្មួញដែលអ្នកលក់ មាន របស់អ្នក ទៅឱ្យ មានឈ្មោះអ្វីខ្លះ?
 Where are they from (Village, Province)?
 តើឈ្មួញទាំងនោះ មកពីទីណាខ្លះ? (ភូមិ, ខេត្ត)
 What is their phone #?
 លេខទូរស័ព្ទឈ្មួញ

[a] Name: _____ [b] _____ [c] _____
 [d] Name: _____ [e] _____ [f] _____
 [g] Name: _____ [h] _____ [i] _____

A19 [a] Since the Khmer New Year, have you sold DUCKS from your home?
 តិចចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក ទៅនឹងផ្ទះ តើអ្នកលក់ ទា ដែរឬទេ?
 1 Yes 0 No **If NO, skip to A24**

[b] How many times?
 តើអ្នកលក់ ទា បានប៉ុន្មានដងហើយ តិចចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក?
 _____ times since Khmer New Year
 ចំនួនលើក

[c] Do you sell DUCKS alive, prepared or both?
 តើលក់បែបណា? រស់, ធ្វើហើយ ឬ ទាំង២យ៉ាង?
 1 Only live 2 Only prepared 3 Both
 លក់តែរស់ លក់តែធ្វើហើយ ទាំង២យ៉ាង

A20 To whom do you sell?
 តើអ្នកលក់ទៅឱ្យអ្នកណាខ្លះ?
 a People in your villager
 អ្នករស់នៅក្នុងភូមិជាមួយគ្នា
 b Middlemen from outside village
 ឈ្មួញមកពីខាងក្រៅភូមិ
 c Middlemen from Vietnam
 ឈ្មួញមកពី វៀតណាម
 d Middlemen from Thailand
 ឈ្មួញមកពី ថៃ
 e Other (specify)
 ផ្សេងទៀត _____

If used a middleman: បើអ្នកលក់ទៅឱ្យ ឈ្មួញដែលដើរទិញតាមផ្ទះ

A21 How many times since the Khmer New Year did you use a middleman to sell poultry [DUCKS]?
 តិចចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក តើអ្នកលក់ ទា របស់អ្នក ទៅឱ្យឈ្មួញដែលដើរទិញ បានប៉ុន្មានដង?
 _____ times a month **If 0, skip to A24**

A22 How many middlemen did you sell to since Khmer New Year?
 តិចចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក តើ អ្នកលក់ ទា របស់អ្នក ទៅឱ្យ ឈ្មួញប៉ុន្មាននាក់?
 _____ middlemen 99 Don't know

A23 What is the name of the middlemen that you use?
 តើឈ្មួញដែលអ្នកលក់ មាន ទា របស់អ្នក ទៅឱ្យ មានឈ្មោះអ្វីខ្លះ?
 Where are they from (Village, Province)?
 តើឈ្មួញទាំងនោះ មកពីទីណាខ្លះ? (ភូមិ, ខេត្ត)
 What is their phone #?
 លេខទូរស័ព្ទឈ្មួញ

[a] Name: _____ [b] _____ [c] _____
 [d] Name: _____ [e] _____ [f] _____
 [g] Name: _____ [h] _____ [i] _____

A24 [a] Do you sell chickens at the market or outside of your village?
 តើអ្នកយក មាន ទៅលក់នៅផ្សារ ឬ នៅក្រៅភូមិរបស់អ្នក ដែរឬទេ?
 1 Yes 0 No **If NO, skip to A26**

[b] How many times since the Khmer New Year did you sell chickens?
 តើអ្នកលក់ បានប៉ុន្មានដងហើយ តិចចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក?
 _____ times since Khmer New Year
 ចំនួនលើក

[c] Do you sell chickens alive, prepared or both?
 តើលក់បែបណា? រស់, ធ្វើហើយ ឬ ទាំង២យ៉ាង?
 1 Only live 2 Only prepared 3 Both
 លក់តែរស់ លក់តែធ្វើហើយ ទាំង២យ៉ាង

A25 During the last 2 months, where did you sell your chickens [market and anywhere else outside the village]?
 ២ ខែចុងក្រោយនេះ តើអ្នកមានលក់ មាន ទៅកន្លែងណាខ្លះ? (អាចជាផ្សារ ឬ កន្លែងដទៃក្រៅភូមិ) **(សូមគូសកន្លែងលក់ [a], [b], [c]... ហើយបំពេញព័ត៌មានក្នុង [1], [2], [3])**

	[1] If Market, Name:	[2] Quantity	[3] Province	[4] District	[5] Village	[6] Distance (km)
a <input type="checkbox"/>	Vietnam					
b <input type="checkbox"/>	Thailand					
c <input type="checkbox"/>	Location 1					
d <input type="checkbox"/>	Location 2					
e <input type="checkbox"/>	Location 3					
f <input type="checkbox"/>	Location 4					
g <input type="checkbox"/>	Location 5					

A26 [a] Do you sell DUCKS at the market or outside of your village? 1 Yes 0 No **If NO, skip to A28**
 តើអ្នកយក ទា ទៅលក់នៅផ្សារ ឬ នៅក្រៅភូមិរបស់អ្នក ដែរឬទេ?
 [b] How many times since the Khmer New Year did you sell ducks? [c] Do you sell chickens alive, prepared or both?
 តើអ្នកលក់ បានប៉ុន្មានដងហើយ គិតចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក? តើលក់បែបណា? រស់, ធ្វើហើយ ឬ ទាំង២យ៉ាង?
 _____ times since Khmer New Year 1 Only live 2 Only prepared 3 Both
 ចំនួនលើកគិតចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក លក់តែរស់ លក់តែធ្វើហើយ ទាំង២យ៉ាង

A27 During the last 2 months, where did you sell your ducks [market and anywhere else outside the village]?
 ២ ខែចុងក្រោយនេះ តើអ្នកមានលក់ ទា ទៅកន្លែងណាខ្លះ? (អាចជាផ្សារ ឬ កន្លែងដទៃក្រៅភូមិ) (សូមគូសកន្លែងលក់ [a], [b], [c]... ហើយបំពេញព័ត៌មានក្នុង [1], [2], [3])

	[1] If Market, Name:	[2] Quantity	[3] Province	[4] District	[5] Village	[6] Distance (km)
a <input type="checkbox"/>	Vietnam					
b <input type="checkbox"/>	Thailand					
c <input type="checkbox"/>	Location 1					
d <input type="checkbox"/>	Location 2					
e <input type="checkbox"/>	Location 3					
f <input type="checkbox"/>	Location 4					
g <input type="checkbox"/>	Location 5					

A28 In general, do you ever sell the whole flock together? ជាទូទៅ តើអ្នកលក់ មានទា អស់ពីទ្រុង ដែរឬទេ ក្នុង១លើកៗ? 1 Yes 0 No
 A29 How long usually is your husbandry for before you sell out the flock? តើអ្នកចិញ្ចឹម មានទា រយៈពេលប៉ុន្មានខែទើបបានលក់ចេញម្តង? _____ months
 A30 Do you sell poultry viscera? តើអ្នកលក់ ត្រឡឹងក្នុងមាន់ទា ដែរឬទេ? 1 Yes 0 No
 A31 Do you sell poultry feathers? តើអ្នកលក់ ស្លាប ឬ រោម មាន់ទា ដែរឬទេ? 1 Yes 0 No

A32 Do you sell poultry's eggs? តើអ្នកលក់ ពងមាន់ ឬ ពងទា ដែរឬទេ? 1 Yes 0 No **If NO, Go to Q# A33**
 A32a Do you wash them before selling? 1 Always wash 2 Sometimes 0 Never
 តើអ្នកមានលាងសំអាត ពងមាន់ ឬ ពងទា ដែរឬទេមុននឹងលក់? លាងរាល់ដង លាងម្តងម្កាល មិនដែលលាងសោះ

Poultry transport practices ការដឹកជញ្ជូនមាន់ទា

Now we are going to ask you some questions about your poultry transporting practices. ខ្ញុំសូមសួរបន្តទាក់ទងនឹងការដឹកជញ្ជូនមាន់ទារបស់អ្នក

A33 Do you transport poultry by vehicle to trade? តើអ្នកមានដឹកជញ្ជូន មាន់ ឬ ទា យកទៅលក់ ដែរឬទេ? 1 Yes 0 No **If NO, Go to Q# B1**
 A34 [a] Chickens ដឹកជញ្ជូន មាន់? 1 Yes 0 No [b] Ducks ដឹកជញ្ជូន ទា? 1 Yes 0 No
 A35 What type of vehicle do you use? តើអ្នកប្រើយានជំនិះអ្វីសំរាប់ដឹកជញ្ជូន? (ចំណើយអាចមានច្រើន, កុំអានចំណើយ)
 a Bicycle កង់ c Own car ឡានផ្ទាល់ខ្លួន e Other (specify) _____
 b Motorbike ម៉ូតូ d Taxi ឡានឈ្នួល **If Taxi, Go to Q#A37** ផ្សេងទៀត
 A36 How often do you wash your vehicle after trade? 1 Everytime 2 Sometimes 0 Never
 តើអ្នកមានលាងសំអាតយានជំនិះរបស់អ្នក រាល់ដង ដែរឬទេពេលត្រលប់ពីលក់វិញ? លាងរាល់ដង លាងម្តងម្កាល មិនដែលលាងសោះ
 A37 What is the longest distance to your trade destination? _____ kilometers
 តើចំងាយផ្លូវឆ្ងាយជាងគេ ដែលអ្នកដឹកមាន់ទាទៅលក់ មានចំងាយប៉ុន្មានគីឡូម៉ែត្រ ពីផ្ទះរបស់អ្នក?
 A38 How many birds can you carry on your vehicle? a Chickens _____ b Ducks _____
 តើអ្នកអាចផ្ទុកមាន់ទាបានប៉ុន្មានក្បាលក្នុងពេលដឹក១លើកៗ? ចំនួនមាន់ ចំនួនទា

A39 Do you use cages to transport birds? តើអ្នកមានប្រើទ្រុង ឬទេក្នុងពេលដឹកជញ្ជូន? 1 Yes 2 No **If No Go to Q# B1**
 A40 How often do you use cages to transport birds? 1 Everytime 2 Sometimes 0 Never
 តើអ្នកមានប្រើទ្រុង រាល់ដង ឬទេ ក្នុងពេលដឹកជញ្ជូនមាន់ទា? រាល់ដង ម្តងម្កាល មិនដែលសោះ
 A41 What are they made of? a Wood b Plastic c Metal d Other (specify) _____
 តើទ្រុងដែលអ្នកប្រើសំរាប់ដឹកជញ្ជូន ធ្វើអំពីអ្វី? ឈើ ប្លាស្ទិក លោហៈ ផ្សេងទៀត **(ចំណើយអាចមានច្រើន)**
 A42 Do you stack cages on your vehicle? 1 Yes 0 No
 តើអ្នក ដាក់ទ្រុងត្រួតលើគ្នា ដែរឬទេនៅពេលអ្នកដឹកមាន់ទាទៅលក់?
 A43 Do you have a tray to catch faecal matter underneath each cage? 1 Yes 0 No
 តើអ្នក មានដាក់ឧបករណ៍អ្វី សំរាប់ត្រងលាមកសត្វដែរឬទេ?
 A44 Do you clean cages used for transport after each time you transport birds? 1 Yes 0 No
 តើអ្នកមានលាងសំអាតទ្រុងរាល់ដង ដែរឬទេ ពេលត្រលប់ពីលក់វិញ?

SECTION B. QUESTIONS ABOUT FIGHTING COCK OWNERSHIP (Head of the family only)

សំណួរទាក់ទងនឹងការចិញ្ចឹម មាន់ជល់

I'm now going to ask you some questions about your fighting cocks.

ខ្ញុំសូមសួរបន្តទាក់ទងនឹងការចិញ្ចឹម មាន់ជល់ របស់អ្នក

B1 How many fighting cocks do you have at home? តើអ្នកមានចិញ្ចឹមមាន់ជល់ប៉ុន្មានក្បាល? | | | | | **If 0, Go to Q#C1**

B2 Did you buy any new fighting cocks within the last 2 months? តើអ្នកមានទិញមាន់ជល់ថ្មីទៀតទេ ក្នុងរយៈពេល ២ខែ ចុងក្រោយនេះ? 1 Yes 0 No **If NO, Go to Q# B5**

B3 How many did you buy? តើអ្នកទិញមាន់ជល់ថ្មីប៉ុន្មានក្បាលថែមទៀត? | | | | |

B4 Where did you buy them from? តើអ្នកទិញមាន់ជល់ថ្មីមកពីកន្លែងណាខ្លះ? [day / month / year] ថ្ងៃ ខែ ឆ្នាំ

a Vietnam [b] When did you buy? ____ / ____ / ____

c Thailand [d] When? ____ / ____ / ____

e Home village [f] When? ____ / ____ / ____

g Other1 (Village/District/Province) _____ [h] When? ____ / ____ / ____

i Other2 (Village/District/Province) _____ [j] When? ____ / ____ / ____

k Other3 (Village/District/Province) _____ [l] When? ____ / ____ / ____

m Other4 (Village/District/Province) _____ [n] When? ____ / ____ / ____

B5 Do you sell fighting cocks? តើអ្នកមានលក់មាន់ជល់ចេញខ្លះទេ? 1 Yes 0 No **If NO, Go to Q# B8**

B6 How many did you sell in the past 2 months? តើអ្នកលក់ចេញបានប៉ុន្មានក្បាល ក្នុងរយៈពេល ២ខែ ចុងក្រោយនេះ? | | | | | ចំនួនក្បាល

B7 Where did you sell them? តើអ្នកលក់ចេញទៅកន្លែងណាខ្លះ?

a Vietnam [d] Other1 (Village/District/Province) _____

b Thailand [e] Other2 (village, district, province) _____

c Home village [f] Other3 (village, district, province) _____

B8 Have any of your fighting cocks been sick since the Khmer New Year? តើមានជល់របស់អ្នក មានជំងឺខ្លះទេ គិតចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក? 1 Yes 0 No **If NO, Go to Q# B12**

B9 What were the symptoms តើមានជល់ដែលជំងឺ ចេញរោគសញ្ញាអ្វីខ្លះ? _____

B10 How many sick fighting cocks died since the Khmer New Year? ក្នុងចំណោមមាន់ជល់ដែលជំងឺ តើមានប៉ុន្មានក្បាលងាប់ គិតចាប់តាំងពីបុណ្យចូលឆ្នាំខ្មែរមក? | | | | | ចំនួនមាន់ជល់ដែលជំងឺងាប់

B11 What do you do with dead cocks? តើអ្នកយកមាន់ជល់ដែលជំងឺងាប់ទៅធ្វើអ្វី? **(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)**

a Eat ធ្វើហូប b Sell លក់ c Bury កប់ចោល d Burn ដុតចោល e Other (specify) _____ ផ្សេងទៀត

B12 Where do you keep your fighting cock? តើអ្នកចិញ្ចឹមមាន់ជល់ដាក់នៅកន្លែងណា? 1 In house 2 Under house 3 In yard 4 Other places _____

ក្នុងផ្ទះ ក្រោម ផ្ទះ ក្នុងទីធ្លាផ្ទះ កន្លែងផ្សេងទៀត

B13 Are they kept separately from other poultry at home? តើអ្នកដាក់មាន់ជល់ ដាច់ដោយឡែកពី មាន់ទា ដទៃដែរឬទេ? 1 Yes 0 No

B14 Do you attend cock fighting events? តើអ្នក លេងជល់មាន់ ដែរឬទេ? 1 Yes 0 No **If NO, Go to Q# C1**

B15 How many times a week do you attend a cock fight? តើអ្នក លេងជល់មាន់ ប៉ុន្មានដងក្នុង១អាទិត្យ? | | | | | Times per week

0 less than once a week

B16 How many different fighting rings cockpits do your cocks fight during the last two months? ក្នុងរយៈពេល ២ខែ ចុងក្រោយនេះ តើអ្នកយកមាន់ទៅជល់នៅប៉ុន្មានសង្កៀន? | | | | | ចំនួនសង្កៀន

B17 Where is your most frequented cockpit? តើសង្កៀនដែលអ្នកចូលចិត្តយកមាន់ទៅជល់ជាងគេនៅណា? 1 Inside village 2 Outside the village

នៅក្នុងភូមិរបស់អ្នក នៅក្រៅភូមិរបស់អ្នក

SECTION C. QUESTIONS TO HOUSEHOLD MEMBERS សំណួរសំរាប់ សមាជិកគ្រួសារ ទូទៅ

Poultry mortality reporting ការផ្តល់ដំណឹងអំពី សត្វមាន់ទាមាស់

I'm now going to ask you some questions about poultry mortality. ខ្ញុំសូមសួររបៀបនៃការកាត់ទងនឹងការផ្តល់ដំណឹងអំពី មាន់ទាមាស់

C1 Have you experience poultry mortality in the since the Khmer New year? 1 Yes 0 No If NO, Go to Q# C7
 តើ មាន់ទា របស់អ្នក មានងាប់ខ្លះទេ គិតចាប់ពីពេលបុណ្យចូលឆ្នាំខ្មែរមក ?

What is the **total number of birds raised** since the Khmer New Year? (the subject may need some time to think about their answer)

C2T តើមាន ចំនួនមាន់ សរុប ដែលបានចិញ្ចឹម ទាំងអស់ប៉ុន្មានក្បាល ចាប់ពីបុណ្យចូលឆ្នាំខ្មែរមក? a Chickens ចំនួនមាន់ សរុប | |

C3T តើមាន ចំនួនទា សរុប ដែលបានចិញ្ចឹម ទាំងអស់ប៉ុន្មានក្បាល ចាប់ពីបុណ្យចូលឆ្នាំខ្មែរមក? b Ducks ចំនួនទា សរុប | |

C4T តើមាន ចំនួនមេមាន់ សរុប ដែលបានចិញ្ចឹម ទាំងអស់ប៉ុន្មានក្បាល ចាប់ពីបុណ្យចូលឆ្នាំខ្មែរមក? c Hens ចំនួនមេមាន់ សរុប | |

Of those, how many of each species were **sick from illness** since the Khmer New Year?

C2S តើមាន មាន់ ដែលឈឺ សរុបទាំងអស់ប៉ុន្មានក្បាល? a Chickens ចំនួនមាន់ ឈឺ | |

C3S តើមាន ទា ដែលឈឺ សរុបទាំងអស់ប៉ុន្មានក្បាល? b Ducks ចំនួនទា ឈឺ | |

C4S តើមាន មេមាន់ ដែលឈឺ សរុបអស់ប៉ុន្មានក្បាល? c Hens ចំនួនមេមាន់ ឈឺ | |

Of those that were sick from illness, how **many died from illness** since the Khmer New Year?

C2 តើមាន មាន់ងាប់ ដោយជំងឺ អស់ប៉ុន្មានក្បាល? a Chickens ចំនួនមាន់ងាប់ | |

C3 តើមាន ទាងាប់ ដោយជំងឺ អស់ប៉ុន្មានក្បាល? b Ducks ចំនួនទាងាប់ | |

C4 តើមាន មេមាន់ងាប់ ដោយជំងឺ អស់ប៉ុន្មានក្បាល? c Hens ចំនួនមេមាន់ងាប់ | |

C5 Did you report poultry's mortality ? 1 Yes 0 No If NO, Go to Q# C7
 តើ អ្នកបានរាយការណ៍ប្រាប់អ្នកណា ដែរឬទេ អំពី មាន់ទា របស់អ្នក ដែលងាប់ ?

C6 To whom do you report? តើ អ្នកបានរាយការណ៍ប្រាប់ទៅអ្នកណាខ្លះ? (ចំណើយអាចមានច្រើន. កុំអានចំណើយ)

a Village chief ប្រធានភូមិ d District vet staff ពេទ្យសត្វស្រុក

b Village health staff / Health center ពេទ្យភូមិ ឬ នៅមណ្ឌលសុខភាព e Provincial vet staff ពេទ្យសត្វខេត្ត

c Village animal health worker / Vet staff ពេទ្យសត្វស្រុកត្រួតត្រាភូមិ ឬ ពេទ្យសត្វភូមិ f Other (specify) ផ្សេងទៀត _____

C7 If there is poultry mortality, what would make you report poultry mortality? (ចំណើយអាចមានច្រើន. កុំអានចំណើយ)

នៅពេលមានមាន់ ទា ងាប់ តើ មានអ្វីខ្លះមិនធម្មតា ដែលធ្វើឱ្យអ្នកទៅរាយការណ៍អំពី មាន់ទា របស់អ្នក ដែលងាប់ ?

a Sudden death c Whole flock die
មាន់ទា ងាប់រហ័សពេក មាន់ទា ទាំងហ្នឹងងាប់អស់រលីង

b Half of flock die d Other: _____
មាន់ទា ងាប់អស់ពាក់កណ្តាលហ្នឹង ផ្សេងទៀត

C8 If there is a wild bird death, would you report it? 1 Yes 0 No
 ប្រសិនបើអ្នកជួបប្រទះ សត្វស្លាប់ព្រៃដែលងាប់ តើអ្នកនឹងទៅរាយការណ៍ ដែរឬទេ ?

Practices when mortality occurred **ការអនុវត្តនៅពេលមាន ម៉ាន់ទាងាប់**

C9 If you encounter poultry mortality in your flocks, what do you do with dead poultry?
 នៅពេលដែលមាន ម៉ាន់ទាងាប់ក្នុងផ្ទះរបស់អ្នក តើអ្នកយក ម៉ាន់ទា របស់អ្នក ដែលងាប់ ទៅធ្វើអ្វីខ្លះ?

(សូមអានចំណើយ)

- [a] Bin បោះចោលនៅចុងសំរាម 1 Yes 0 No
- [b] Bury កប់ចោល 1 Yes 0 No **If no, skip to d**
- [c] Can you point out where you buried them? [Check **Yes** only when they can show you where they buried poultry] 1 Yes 0 No
 សូមបង្ហាញកន្លែងដែលអ្នកបានកប់ [ត្រូវ YES នៅពេលណាដែលគេអាចបង្ហាញកន្លែងកប់]
- [d] Burn ដុតចោល 1 Yes 0 No **If no, skip to f**
- [e] Can you point out where you burned them? [Check **Yes** only when they can show you where they burned poultry] 1 Yes 0 No
 សូមបង្ហាញកន្លែងដែលអ្នកបានដុត [ត្រូវ YES នៅពេលណាដែលគេអាចបង្ហាញកន្លែងដុត]
- [f] Feed other animals បោះអោយសត្វផ្សេងស៊ី 1 Yes 0 No
- [g] Prepare for selling ធ្វើលក់ 1 Yes 0 No **If no, skip to i**
- [h] Where did you sell? តើអ្នក យកទៅលក់ នៅឯណា? (Village, Commune, District, Province) _____
- [i] Prepare for food យកមកចម្អិនធ្វើម្ហូប 1 Yes 0 No
- [j] Sell carcass លក់ម៉ាន់ទាងាប់ទាំងមូល 1 Yes 0 No **If no, skip to l**
- [k] Where did you sell? តើអ្នក យកទៅលក់ នៅឯណា? (Village, Commune, District, Province) _____
- [l] Give away to neighbor ឱ្យទៅអ្នកជិតខាង 1 Yes 0 No
- [m] Throw into water sources បោះចោលទៅក្នុងទឹក 1 Yes 0 No
- [n] Throw away ត្រូវវែងចោល 1 Yes 0 No
- [o] Other (specify) ផ្សេងទៀត _____

C10 What do you do if/when you have **sick poultry** in your flocks ?

នៅពេលដែលមាន ម៉ាន់ទាលី ក្នុងផ្ទះរបស់អ្នក តើអ្នកយក ម៉ាន់ទា របស់អ្នក ដែលលី ទៅធ្វើអ្វីខ្លះ?

(សូមអានចំណើយ)

Would you:

- [a] Quarantine separately from other in the flocks? ដាក់ ម៉ាន់ទា ដែលលី ដាច់ដោយឡែកពី ម៉ាន់ទាដទៃទៀត 1 Yes 0 No
- [b] Slaughter for selling? សំលាប់យកសាច់លក់ 1 Yes 0 No **If no, skip to e**
- [c] Where did you sell? តើអ្នកយកទៅលក់នៅឯណា? (village, commune, district, Province) _____
- [d] Did you use a middleman? តើអ្នកមានលក់ទៅឱ្យឈ្មួញដើរទិញម៉ាន់ទាដែរឬទេ? 1 Yes 0 No
- [e] Slaughter for food? សំលាប់យកសាច់ធ្វើម្ហូប 1 Yes 0 No
- [f] Give to neighbors for consumption? ឱ្យទៅអ្នកជិតខាងយកទៅធ្វើម្ហូប 1 Yes 0 No
- [g] Sell alive? លក់ទាំងរស់ 1 Yes 0 No **If no, skip to j**
- [h] Where did you sell? តើអ្នកយកទៅលក់នៅឯណា? (village, commune, district, Province) _____
- [i] Did you use a middleman? តើអ្នកមានលក់ទៅឱ្យឈ្មួញដើរទិញម៉ាន់ទាដែរឬទេ? 1 Yes 0 No
- [j] Do nothing? មិនបានធ្វើអ្វីទាំងអស់ 1 Yes 0 No
- [k] Other (specify) ផ្សេងទៀត _____

C11 What do you do with **remaining poultry that survived** in the flock ?

តើអ្នកយក មាន់ទា របស់អ្នក ដែលនៅសល់ពីលី ឬ ងាប់ ទៅធ្វើអ្វីខ្លះ?

(សូមអានចំណើយ)

Would you:

- [a] Quarantine separately from other in the flocks? ដាក់ មាន់ទា ដែលលី ដាច់ដោយឡែកពី មាន់ទាដទៃទៀត 1 Yes 0 No
- [b] Slaughter for selling? សំលាប់យកសាច់លក់ 1 Yes 0 No **If no, skip to e**
- [c] Where did you sell? តើអ្នកយកទៅលក់នៅឯណា? (village, commune, district, Province) _____
- [d] Did you use a middleman? តើអ្នកមានលក់ទៅឱ្យឈ្មួញដើរទិញមាន់ទាដែរឬទេ? 1 Yes 0 No
- [e] Slaughter for food? សំលាប់យកសាច់ធ្វើម្ហូប 1 Yes 0 No
- [f] Give to neighbors for consumption? ឱ្យទៅអ្នកជិតខាងយកទៅធ្វើម្ហូប 1 Yes 0 No
- [g] Sell alive? លក់ទាំងរស់ 1 Yes 0 No **If no, skip to j**
- [h] Where did you sell? តើអ្នកយកទៅលក់នៅឯណា? (village, commune, district, Province) _____
- [i] Did you use a middleman? តើអ្នកមានលក់ទៅឱ្យឈ្មួញដើរទិញមាន់ទាដែរឬទេ? 1 Yes 0 No
- [j] Do nothing? មិនបានធ្វើអ្វីទាំងអស់ 1 Yes 0 No
- [k] Other (specify) ផ្សេងទៀត _____

Knowledge and attitude ចំណេះដឹង និង ឥរិយាបថ

- C12 Do you think it's important to report poultry mortality? 1 Yes **If YES, Go to Q# C12b**
- តើអ្នកគិតថាការរាយការណ៍ អំពី មាន់ទា ងាប់ មានសារៈសំខាន់ដែរឬទេ? 0 No **If NO, Go to Q# C12a**
- 9 Don't know **If DK, Go to Q# C13**

- C12a **If NO,** Why is it not important? ហេតុអ្វីបានជាអ្នកគិតថាការរាយការណ៍ អំពី មាន់ទា ងាប់ **មិនមាន** សារៈសំខាន់? **(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)**
- a No help from vet staff or authorities ព្រោះមិនមានជំនួយពីពេទ្យសត្វ ឬ អាជ្ញាធរ d Don't know មិនដឹង
- b Too few death ព្រោះមាន់ទាងាប់តិចតួច e Other (specify) ផ្សេងទៀត
- c Poultry are not important (like pigs or cattles..) ព្រោះមាន់ទាមិនសំខាន់ដូចសត្វគោក្របីទេ

- C12b **If YES,** Why is it important? ហេតុអ្វីបានជាអ្នកគិតថាការរាយការណ៍ អំពី មាន់ទា ងាប់ **មាន សារៈសំខាន់**? **(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)**
- a It could be AI ព្រោះអាចមានជំងឺគ្រុនផ្តាសាយបក្សី c Don't know មិនដឹង
- b We can have advices from VAHWs ព្រោះគិតថាអាចទទួលបានការណែនាំពីបុគ្គលិកពេទ្យសត្វ d Other (specify) ផ្សេងទៀត

C13 If you report mortality, what do you expect in return? បើអ្នករាយការណ៍ តើអ្នកចង់បានអ្វីខ្លះត្រលប់មកវិញ?

C13a And from whom? តើអ្នកចង់បានពីអ្នកណា? _____

- C14 What do you think discourages villagers to report poultry mortality? **(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)**
- តាមយោបល់របស់អ្នក តើអ្វីខ្លះដែលធ្វើឱ្យអ្នកភូមិមិនចង់ទៅរាយការណ៍ អំពីមាន់ទាដែលងាប់?
- a Fear of poultry culling ខ្លាចគេមកសំលាប់មាន់ទារបស់ពួកគេអស់ c Fear of panic in the village ខ្លាចធ្វើឱ្យមានការភ័យជ្រួលច្របល់នៅក្នុងភូមិ
- b Problems with selling ខ្លាចលក់មាន់ទាលែងបាន d Don't know មិនដឹង
- e Don't know where/to whom to report មិនសឹងទៅប្រាប់អ្នកណា, នៅទីណា
- f Poultry are not important like cattle មាន់ទាជាសត្វតូចតាច មិនសំខាន់ដូចគោ ក្របីទេ
- g Other? (specify) ផ្សេងទៀត _____

- C15 What, do you think, would help encourage farmers to report poultry's mortality ? **(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)**
- តាមយោបល់របស់អ្នក តើអ្វីខ្លះដែលអាចជួយលើកទឹកចិត្តអ្នកភូមិឱ្យ ទៅរាយការណ៍ អំពីមាន់ទាដែលងាប់?
- a Awareness / education on the risk of AI ការអប់រំអំពីគ្រោះថ្នាក់បណ្តាលមកពីជំងឺគ្រុនផ្តាសាយបក្សី d Incentive ការផ្តល់ជំរាញដល់អ្នកទៅរាយការណ៍
- b Help from authorities (drug, food, training...) ជំនួយពីអាជ្ញាធរ (ឧ: ថ្នាំ, ចំណីសត្វ, វគ្គបណ្តុះបណ្តាល...) e Buying sick or dead poultry at lower price ជួយទិញមាន់ទាដែល លី ឬ ងាប់ ដោយតម្លៃថោកៗ
- c Compensation in case of culling មានជាសំណងក្នុងករណីសំលាប់មាន់ទា f Don't know មិនដឹង
- g Ask them to report ស្នើសុំឱ្យគាត់ទៅរាយការណ៍
- h Other (specify) ផ្សេងទៀត _____

C16 what do you think authorities would do if you reported mortality in your flocks?

តើគិតថាអាជ្ញាធរគួរធ្វើអ្វីខ្លះ នៅពេលដែលអ្នក ទៅរាយការណ៍ អំពីមាត់ទាដែលងាប់?

C17 If your poultry were culled, what do you expect?

ក្នុងករណី មានការបាត់បង់អ្នកត្រូវសំលាប់ តើអ្នកគិតចង់បានអ្វីខ្លះជាសំណងត្រឡប់មកវិញ?

(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)

- a Replace poultry សងជា មាត់ទា មកវិញ
- b Money back សងជា លុយ មកវិញ
- c Don't know មិនដឹង
- d Nothing មិនចង់បានអ្វីទាំងអស់
- e Other(specify) ផ្សេងទៀត _____

PRACTICES ការអនុវត្ត

- C18 Do you attend cock fighting events? តើអ្នកទៅលេង ឬ មើលជល់មាត់ដែរឬទេ? 1 Yes 0 No
- C19 Do you care or help care for fighting cocks? តើអ្នកមើលថែទាំ សំអាតមាត់ជល់ (ឬ ជួយមើលថែទាំមាត់ជល់) ដែរឬទេ? 1 Yes 0 No **If NO, Go to Q# C21**
- C19a How many days per week you take care of your cocks? តើអ្នកមើលថែទាំ សំអាតមាត់ជល់ ប៉ុន្មានថ្ងៃ ក្នុង១អាទិត្យ? _____ times per week
- C19b Do you wear gloves or masks when caring your cocks? តើអ្នកមានពាក់ស្រោមដៃ ឬ ម៉ាស់ ដែរឬទេ នៅពេលដែលអ្នកមើលថែទាំ សំអាតមាត់ជល់? 1 Always 2 Sometimes 0 <1 time a week 0 Never
ពាក់ជានិច្ច ពាក់ម្តងម្កាល មិនដែលពាក់សោះ
- C20a Do you: Touch bloody cocks? តើអ្នកមានប៉ះពាល់មាត់ជល់ដែលមានហូរឈាម ដែរឬទេ? 1 Yes 0 No
- C20b Blow the beak? តើអ្នកមាន ផ្លុំ ចំពុះមាត់ជល់ ដែរឬទេ? 1 Yes 0 No
- C20c Kiss, Suck or lick wounds? តើអ្នកមាន បិតរបូសមាត់ជល់ ដែរឬទេ? 1 Yes 0 No
- C20d Share water from the same bottle used for drinking or spraying? តើអ្នកមាន ប្រើទឹករួមជាមួយមាត់ជល់ ដែរឬទេ? 1 Yes 0 No
- C20e Clean the trachea by swab or feather? តើអ្នកមាន សំអាតបំពង់ក មាត់ជល់ ដោយយកសំឡី ឬ ស្លាបត្បាញ ដែរឬទេ? 1 Yes 0 No
- C20f Clean feathers? តើអ្នកមាន សំអាតស្លាប និង រោម ឱ្យមាត់ជល់ ដែរឬទេ? 1 Yes 0 No

Practices ការអនុវត្ត

I'm now going to ask you some questions about your daily life. **ខ្ញុំសូមសួរបន្តទាក់ទងនឹង ជីវិតប្រចាំថ្ងៃរបស់អ្នក**

- C21 Do you touch sick or dead poultry with bare hands? តើអ្នក ប៉ះពាល់ មាត់ទាលី ឬ ងាប់ ដោយដៃទទេ ដែរឬទេ? 1 Yes 0 No
- C22 Do children in your household play (touch and catch) with poultry? តើក្មេងៗនៅក្នុងផ្ទះរបស់អ្នក មានប៉ះពាល់ ឬ លេង ជាមួយមាត់ទា ដែរឬទេ? 1 Yes 0 No 9 DK
- C23 Do your children swim in ducks ponds? តើកូនរបស់អ្នក ទៅមុជ ហែលទឹកលេង ក្នុងស្រះដែលមានទាចុះ ដែរឬទេ? 1 Yes 0 No 9 DK
- C23a Do you swim or fish in water (ponds ...) where poultry have access? តើអ្នក មុជទឹក ឬ លេងទឹក ឬ បាចរកត្រី ក្នុងទឹក (ឧ. ស្រះ, ត្រពាំង ឬ បឹង ...) ដែលមានទាចុះ ដែរឬទេ? 1 Yes 0 No 9 DK
- C24 Do you take dead chicken or poultry from yard for food? តើអ្នកមានរើសយក មាត់ទាដែលងាប់ យកមកធ្វើម្ហូប ដែរឬទេ? 1 Yes 0 No
- C24a Do you remove feathers from sick poultry? តើអ្នក បោចរោម មាត់ទាដែលឈឺ (ធ្វើម្ហូប...) ដែរឬទេ? 1 Yes 0 No
- C25 Do you take dead wild birds from field for food? តើអ្នកមានរើសយក សត្វព្រៃដែលងាប់តាមវាល យកមកធ្វើម្ហូប ដែរឬទេ? 1 Yes 0 No
- C26 Do you eat wild birds? តើអ្នកហូបសាច់សត្វព្រៃដែរឬទេ? 1 Yes 0 No
- C27 In your family, are you responsible for going to the market (buying food)? តើអ្នកទៅផ្សារទិញម្ហូប ដែរឬទេ? 1 Yes 0 No
- C27a Have you ever bought poultry from the market for food? តើអ្នកមានទិញមាត់ទាពីផ្សារ មកធ្វើម្ហូប ដែរឬទេ? 1 Yes 0 No

28	Do you cook or help in cooking poultry for your family ? តើអ្នក ធ្វើម្ហូប ឬ ជួយធ្វើម្ហូប ដែរឬទេ?	1 <input type="checkbox"/> Everyday រាល់ថ្ងៃ	2 <input type="checkbox"/> Sometimes ម្តងម្កាល	0 <input type="checkbox"/> Never មិនដែលសោះ
29	Do you: Boil birds? តើអ្នក ស្ទោរចំអិនមាន់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
3a	Slaughter/ bleed birds? តើអ្នក អារក សំលាប់ មាន់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
3b	Cut meat? តើអ្នក កាប់,ហាន់ សាច់មាន់ សាច់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
3c	Wash meat? តើអ្នក លាង សាច់មាន់ សាច់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
3d	Remove internal organs from birds? តើអ្នក រះយកគ្រឿងក្នុង មាន់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
3e	Wash internal organs? តើអ្នក លាង គ្រឿងក្នុង មាន់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
3f	Do you eat raw or half cooked chicken eggs (eggs with runny yokes)? តើអ្នក ហូប ឬ ហុត ពងមាន់នៅ ឬ ចំអិន នៅជីវព្នៅ ដែរឬទេ ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
3g	Do you eat raw chicken meat? តើអ្នក ហូប ម្ហូបដែលធ្វើពី សាច់មាន់ នៅ ដែរឬទេ ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
3h	Do you eat chicken meat that is pink in color (has "pink spots")? តើអ្នក ហូប សាច់មាន់ ដែលចំអិន នៅនៅ ក្រហមដុំៗ ដែរឬទេ ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
3i	Do you eat raw or half cooked duck eggs (eggs with runny yokes)? តើអ្នក ហូប ឬ ហុត ពងទា នៅ ឬ ចំអិន នៅជីវព្នៅ ដែរឬទេ ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
3j	Do you eat raw duck meat? តើអ្នក ហូប ម្ហូបដែលធ្វើពីសាច់ទា នៅ ដែរឬទេ ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
3k	Do you eat duck meat that is pink in color (has "pink spots")? តើអ្នក ហូប សាច់ទា ដែលចំអិន នៅនៅ ក្រហមដុំៗ ដែរឬទេ ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
30	Have you ever <u>prepared</u> wild birds for food? តើអ្នកធ្លាប់ ធ្វើ,កាប់ សាច់សត្វស្លាបព្រៃចំអិនធ្វើម្ហូប ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
31	Do you prepare poultry near pond, river, water well? តើអ្នក មានយកមាន់ទៅធ្វើ នៅក្បែរ ត្រពាំង ព្រែក ទន្លេ ឬ អណ្តូង ដែរឬទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> No pond/river/ open water well មិនមាន ត្រពាំង ព្រែក ទន្លេ ឬ អណ្តូង
32	Do wash poultry products directly in the water source (pond/river)? តើអ្នក មានយកមាន់ទៅ លាងផ្ទាល់ នៅក្នុង ត្រពាំង ព្រែក ឬ ទន្លេ ដែរឬទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> No pond/river មិនមាន ត្រពាំង ព្រែក ឬ ទន្លេ
33	Do you care or help care for poultry? តើអ្នក មានជួយមើលថែទាំ មាន់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No If NO, go to Q35
34a	How many times a week do you clean your poultry cages/areas ? តើអ្នក សំអាត ទ្រុងមាន់ទា ប៉ុន្មានដងក្នុង១អាទិត្យ?			_____ times per week 0 <input type="checkbox"/> <1 time a week
34b	When caring for poultry: នៅពេលដែលអ្នក ថែទាំ សំអាត ទ្រុងមាន់ទា			
34c	Do you wear plastic bags over hands? តើអ្នក មានពាក់ថង់ប្លាស្ទិក ការពារនៅដៃ ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
34d	Do you have any gloves in your household? តើអ្នក មានស្រោមដៃដែរ ដែរឬទេ នៅក្នុងផ្ទះរបស់អ្នក?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No If NO, Go to C34d
34e	How often do wear gloves when touching poultry? តើអ្នក ពាក់ស្រោមដៃ រាល់ដងដែរឬទេ នៅពេលប៉ះពាល់មាន់ទា?	1 <input type="checkbox"/> Everyday	2 <input type="checkbox"/> Sometimes	0 <input type="checkbox"/> Never មិនដែលពាក់សោះ
34f	Do you have any rubber boots in your household? តើអ្នក មានស្បែកជើងជ័រ ក វែង ដែរឬទេ នៅក្នុងផ្ទះ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No If NO, Go to C34f
34g	How often do you wear rubber boots when caring for poultry? តើអ្នក ពាក់ស្បែកជើងជ័រ ក វែង រាល់ដងដែរឬទេ នៅពេលធ្វើការសំអាតទ្រុងមាន់ទា?	1 <input type="checkbox"/> Everyday	2 <input type="checkbox"/> Sometimes	0 <input type="checkbox"/> Never មិនដែលពាក់សោះ

4f Do you have any aprons in your household? តើអ្នក មានអាវកំពារ ដែរឬទេ នៅក្នុងផ្ទះរបស់អ្នក? 1 Yes 0 No **If NO, Go to C34h**

g How often do you wear an apron when caring for poultry? 1 Everyday 2 Sometimes 0 Never
 តើអ្នក ពាក់អាវកំពារ រាល់ដងដែរឬទេ នៅពេលបិទពាល់ ថែទាំ មាន់ទា? ពាក់រាល់ដង ពាក់ម្តងម្កាល មិនដែលពាក់សោះ

h Do you have any masks in you household? តើអ្នក មានម៉ាស់ (របាំងមុខ) ដែរឬទេ នៅក្នុងផ្ទះរបស់អ្នក? 1 Yes 0 No **If NO, Go to C35**

4i How often do you wear face mask when caring for poultry? 1 Everyday 2 Sometimes 0 Never
 តើអ្នក ពាក់ម៉ាស់ (របាំងមុខ) ការពារ រាល់ដងដែរឬទេ នៅពេលបិទពាល់ ថែទាំ មាន់ទា? ពាក់រាល់ដង ពាក់ម្តងម្កាល មិនដែលពាក់សោះ

5 Do you, personally, feed your poultry ? តើអ្នក ឱ្យចំណី មាន់ទា ដោយខ្លួនឯង ឬទេ? 1 Yes 0 No **If NO, Go to C36**

a How many times a week do you, personally, feed poultry? តើអ្នក ឱ្យចំណី មាន់ទា ប៉ុន្មានដង ក្នុង១អាទិត្យ? _____ Times 0 <1 time a week

6 Do your poultry go in rice fields or lake ? តើមាន់ទារបស់អ្នកចេញដើរកសិ តាមវាលស្រែ ឬ បឹង ដែរឬទេ? 1 Yes 0 No

7 Do you use poultry faeces for manure ? តើអ្នកប្រើលាមកមាន់ទា សំរាប់ធ្វើដី ដែរឬទេ? 1 Yes 0 No

8 Have you ever been to Vietnam ? តើអ្នក ធ្លាប់ទៅដល់ប្រទេសវៀតណាម ដែរឬទេ? 1 Yes 0 No **If NO, Go to C39**

a How many times per month do you go to Vietman? តើអ្នក ទៅប៉ុន្មានដង ក្នុង១ខែ? _____ times per month 0 <1 time a month

b Do you go there for trade (sell or buy) poultry or poultry products? អ្នក ទៅដើម្បីលក់ , ទិញ មាន់ទា តើមែនឬទេ? 1 Yes 0 No **If no, go to C39**

c Which provinces do you go to sell/buy? អ្នក ទៅដល់ខេត្តណាខ្លះ? _____

9 Have you ever been to Thailand ? តើអ្នក ធ្លាប់ទៅដល់ប្រទេសថៃ ដែរឬទេ? 1 Yes 0 No **If NO, Go to C40s**

a How many times per month do you go to Thailand? តើអ្នក ទៅប៉ុន្មានដង ក្នុង១ខែ? _____ times per month 0 <1 time a month

b Do you go there for trade (sell or buy) poultry or poultry products? អ្នក ទៅដើម្បីលក់ , ទិញ មាន់ទា តើមែនឬទេ? 1 Yes 0 No **If no, go to C40s**

c Which provinces do you go to sell/buy? អ្នក ទៅដល់ខេត្តណាខ្លះ? _____

c hygiene practices ការអនុវត្ត អនាម័យជាមូលដ្ឋាន

s Do you have soap in your household? តើ អ្នកមានសាប៊ូ ឬទេ នៅក្នុងផ្ទះរបស់អ្នក ? 1 Yes 0 No

0 Do you wash your hands with soap after you touch poultry? 1 Yes 0 No **If NO, Go to C40b**
 តើអ្នក លាងដៃជាមួយសាប៊ូ ដែរឬទេ ក្រោយពេលបិទពាល់មាន់ទា?

a How often? តើអ្នក លាង ជាមួយសាប៊ូ រាល់ដងដែរឬទេ? 1 Everytime 2 Sometimes 3 Rarely

b If you have no soap or run out of soap, what do you use instead of soap to wash your hand?
 បើអ្នកគ្មានសាប៊ូ តើអ្នក ប្រើអ្វីជំនួស សំរាប់លាងដៃ? _____

1 Which water source do you usually use in your household? តើអ្នក យកទឹកអ្វី មកពីណា សំរាប់ប្រើប្រាស់ក្នុងគ្រួសារ? **(ចំណើយអាចមានច្រើន)**

a Open water well ទឹកអណ្តូងយោង c Ponds ទឹកត្រពាំង, ស្រះ e Lake ទឹកបឹង g Other (specify) _____ ផ្សេងទៀត

b Water well with pump ទឹកអណ្តូងស្តាប់ d Water tap ទឹកម៉ាស៊ីន f River ទឹកទន្លេ

C42 If pond or lake or river does, poultry has access to those water sources as well? 1 [] Yes 0 [] No 9 [] Don't Know
បើ ត្រពាំង បឹង ឬ ទន្លេ តើ មាន ម្ចាស់ ចុះទៅក្នុងទឹកនោះដែរឬទេ?

C42a Which water source do you used to bathe? តើអ្នក ងូតទឹកអ្វី នៅឯណា ? (ចំណើយអាចមានច្រើន)
a [] Open water well ទឹកអណ្តូងយោង c [] Ponds ទឹកត្រពាំង, ស្រះ e [] Lake ទឹកបឹង g [] Other (specify) ផ្សេងទៀត
b [] Water well with pump ទឹកអណ្តូងស្នប់ d [] Water tap ទឹកម៉ាស៊ីន f [] River ទឹកទន្លេ

C42b If pond or lake or river, do poultry have access to these water sources? 1 [] Yes 0 [] No 9 [] Don't Know
បើ ត្រពាំង បឹង ឬ ទន្លេ តើ មាន ម្ចាស់ ចុះទៅក្នុងទឹកនោះដែរឬទេ?

Knowledge about AI / bird flu ចំណេះដឹងអំពី ជំងឺត្រូវផ្តាសាយបក្សី
I'm now going to ask you some questions about avian influenza. ខ្ញុំសូមសួររប្អូនទាក់ទងនឹង ជំងឺត្រូវផ្តាសាយបក្សី

C43 Have you heard of AI / bird flu? តើអ្នកធ្លាប់ឮអំពី ជំងឺត្រូវផ្តាសាយបក្សី ដែរឬទេ? 1 [] Yes 0 [] No If NO, Go to C47

C44 Where did you hear that information from? តើអ្នកធ្លាប់ឮអំពី ជំងឺត្រូវផ្តាសាយបក្សី មកពីណាខ្លះ? (ចំណើយអាចមានច្រើន, កុំអានចំណើយ)
a [] Village vet staffs ពេទ្យសត្វភូមិ d [] Radio វិទ្យុ g [] Public poster ផ្ទាំងរូបភាពអប់រំតាមភូមិ j [] Someone in the family សមាជិតគ្រួសារ
b [] Health staff / health center ពេទ្យភូមិ ឬ នៅមណ្ឌលសុខភាព e [] Television ទូរទស្សន៍ h [] Brochures កូនសៀវភៅអប់រំតូចៗ k [] Other (specify) ផ្សេងទៀត
c [] Village chiefs ប្រធានភូមិ f [] Newspaper កាសែត i [] NGO health education sessions វគ្គអប់រំរបស់អង្គការនានា

C44 What would you consider to be your MAIN source of AI information? (open question) ក្នុងចំណោមប្រភពទាំងនោះ តើប្រភពមួយណា ដែលអ្នកគិតថា បានផ្តល់ព័ត៌មានអំពី ផ្តាសាយបក្សី ដល់អ្នក បានច្រើនជាងគេ ?

C45 Have you taught your children about AI តើអ្នកធ្លាប់បង្រៀន កូនៗអ្នក ឬ ក្មេងៗ អំពីជំងឺត្រូវផ្តាសាយបក្សី ដែរឬទេ? 1 [] Yes 0 [] No If NO, Go to C46

C45a What have you taught them (open question) តើអ្នកប្រាប់ កូនៗអ្នក យ៉ាងដូចម្តេចខ្លះ?

C46 In your own words, what is bird flu ? តាមយោបល់របស់អ្នក តើ ជំងឺត្រូវផ្តាសាយបក្សី ជាអ្វី? (ចំណើយអាចមានច្រើន, កុំអានចំណើយ)
a [] Poultry disease (duck/chicken...) ជំងឺម្ចាស់ ទា c [] Human disease ជំងឺមនុស្ស e [] Pig disease ជំងឺជ្រូក g [] Can be transmitted to human អាចឆ្លងមកមនុស្សបាន
b [] Can kill poultry អាចសំលាប់ ម្ចាស់ ទា បាន d [] Can kill human អាចសំលាប់ មនុស្ស បាន f [] Wild bird disease ជំងឺសត្វស្លាបព្រៃ h [] Other (specify) ផ្សេងទៀត

C47 Could you tell if your flock were infected by AI? តើអ្នកគិតថា ម្ចាស់ទាររបស់អ្នក ធ្លាប់មានឆ្លងជំងឺត្រូវផ្តាសាយបក្សី ពីមុនមក ដែរឬទេ? 1 [] Yes 0 [] No 9 [] Don't know

C48 Do you think your poultry are at risk of being infected with AI? តើអ្នកគិតថាម្ចាស់ទាររបស់អ្នកអាចឆ្លងជំងឺត្រូវផ្តាសាយបក្សី ដែរឬទេ? 1 [] Yes 0 [] No 9 [] Don't know

C54 Can humans get AI? តើមនុស្សអាចឆ្លងជំងឺគ្រុនផ្តាសាយបក្សីដែរឬទេ? 1 [] Yes 0 [] No 9 [] Don't know **If No or Don't know, skip to C56**

C55 How can humans catch AI? តើជំងឺគ្រុនផ្តាសាយបក្សីឆ្លងមកមនុស្ស ដោយរបៀបណា? **(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)**
a [] Contact with sick/dead poultry ការប៉ះពាល់មាន់ទាស្លាប់
b [] Contact with poultry feces ការប៉ះពាល់អាចម៍សត្វ
c [] Eating undercooked poultry products បរិភោគមាន់ទាមិនទាន់ឆ្អិនល្អ " សាច់ ស៊ុត .."
d [] Eating raw poultry products (eggs / blood pudding) បរិភោគឆ្មៅ " ស៊ុត សាច់ ឈាម .."
e [] Contact with contaminated farm equipment ការប៉ះពាល់សំភារៈកសិដ្ឋានមានផ្ទុកមេរោគ
f [] Don't know មិនដឹង
g [] Other (Specify) ផ្សេងទៀត _____

C56 Do you think Bird flu can kill (is fatal for) humans? តើអ្នកគិតថា ជំងឺគ្រុនផ្តាសាយបក្សី អាចសំលាប់មនុស្សបាន ដែរឬទេ? 1 [] Yes 0 [] No 9 [] Don't know

C57 In Cambodia, Have there been any fatal human cases of AI? តើមានមនុស្សស្លាប់ដោយ ជំងឺគ្រុនផ្តាសាយបក្សី ដែរឬទេ នៅប្រទេសកម្ពុជាយើង? 1 [] Yes 0 [] No 9 [] Don't know

C58 Do you think AI can be transmitted to **HUMANS** by... តើអ្នកគិតថា ជំងឺគ្រុនផ្តាសាយបក្សីឆ្លងមកមនុស្ស តាម...
[a] Eating well cooked poultry's products ? ហូបសាច់ ស៊ុត មាន់ទាដែលបានចំអិនឆ្អិនល្អ? 1 [] Yes 0 [] No 9 [] Don't know
[b] Eating undercooked poultry's products ? ហូបសាច់ ស៊ុត មាន់ទាដែល ចំអិននៅឡើយ មិនឆ្អិនល្អ? 1 [] Yes 0 [] No 9 [] Don't know
[c] Eating poultry eggs from healthy chickens? ហូបពងមាន់ ពងទា? 1 [] Yes 0 [] No 9 [] Don't know
[d] Eating poultry bought from market? ហូបសាច់មាន់ទា ដែលទិញយកមកពីផ្សារ? 1 [] Yes 0 [] No 9 [] Don't know
[e] Eating your own poultry prepared at home? ហូបសាច់មាន់ទា ដែលចិញ្ចឹម និង ធ្វើ ដោយខ្លួនឯង? 1 [] Yes 0 [] No 9 [] Don't know
[f] Eating poultry imported from outside Cambodia? ហូបសាច់មាន់ទា ដែលនាំចូលមកពីក្រៅប្រទេសកម្ពុជា? 1 [] Yes 0 [] No 9 [] Don't know
[g] Swimming in ponds? មុជទឹកក្នុងស្រះ 1 [] Yes 0 [] No 9 [] Don't know
[h] Touching wild birds ? ប៉ះពាល់សត្វស្លាបព្រៃ? 1 [] Yes 0 [] No 9 [] Don't know
[i] Eating wild birds? ហូបសាច់សត្វស្លាបព្រៃ? 1 [] Yes 0 [] No 9 [] Don't know
[j] From other people? ពីមនុស្សម្នាក់ ទៅ ម្នាក់ទៀត 1 [] Yes 0 [] No 9 [] Don't know
[k] Touching healthy poultry ? ប៉ះពាល់សត្វមាន់ទា អត់មានជំងឺ? 1 [] Yes 0 [] No 9 [] Don't know
[l] Touching sick or dead poultry with bare hands? ប៉ះពាល់សត្វមាន់ទា ដែលឈឺ ងាប់ ដោយដៃទទេ? 1 [] Yes 0 [] No 9 [] Don't know
[m] Touching poultry faeces? ប៉ះពាល់ជាមួយឈាមសត្វមាន់ទា? 1 [] Yes 0 [] No 9 [] Don't know
[n] From sexual contact? តាមការរួមភេទ 1 [] Yes 0 [] No 9 [] Don't know
[o] Touching poultry blood? ប៉ះពាល់ជាមួយ ឈាមសត្វមាន់ទា? 1 [] Yes 0 [] No 9 [] Don't know
[p] Slaughtering poultry ? សំលាប់មាន់ទាធ្វើម្ហូប? 1 [] Yes 0 [] No 9 [] Don't know
[q] Using the same cutting board for poultry and food products? ប្រើជ្រុងតែ១ រួម សំលាប់ សាច់មាន់ ទា និង បន្លែ សាច់ គ្រប់មុខ ដីទៃទៀត? 1 [] Yes 0 [] No 9 [] Don't know

C59 Do you think AI can be transmitted to CHILDREN by touching / playing with sick / dead poultry (with bare hands)? តើអ្នកគិតថា កូនក្មេង អាចឆ្លងជំងឺគ្រុនផ្តាសាយបក្សី ដោយ ប៉ះពាល់ ឬ លេង ជាមួយ មាន់ទាដែល ឈឺ ឬ ងាប់ ដែរឬទេ? 1 [] Yes 0 [] No 9 [] Don't know

C60 Do you know any AI / Bird Flu signs and symptoms in **HUMANS**? តើអ្នកស្គាល់រោគសញ្ញាជំងឺគ្រុនផ្តាសាយបក្សី នៅក្នុងខ្លួន **មនុស្ស** ដែរឬទេ? 1 [] Yes 0 [] No **If NO, Go to C61**

C60a What are those signs and symptoms? ប្រសិនបើដឹង, តើរោគសញ្ញាទាំងនោះមានអ្វីខ្លះ? **(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)**
a [] Difficult/fast breathing ពិបាកដកដង្ហើម
b [] Fever គ្រុនក្តៅ
c [] Cough ក្អក
d [] Muscle Ache ឈឺសាច់ដុំ
e [] Sore throat ឈឺករ
f [] Other (Specify) ផ្សេងទៀត _____

C61 Do you believe you can protect your poultry and yourself from AI? 1 Yes 0 No 9 Don't know
 តើអ្នកជឿថា អ្នកអាចការពារ មាន់ទា របស់អ្នក និង ខ្លួនអ្នកផ្ទាល់ មិនឱ្យឆ្លងជំងឺគ្រុនផ្តាសាយបក្សី បាន ដែរឬទេ?

C62 Do you think touching sick/dead poultry can cause fever? 1 Yes 0 No
 តើអ្នកគិតថា ការប៉ះពាល់ មាន់ទា ដែលឈឺ ឬ ងាប់ដោយសារជំងឺ អាចបង្កអោយកើតគ្រុនក្តៅ ដែរឬទេ?

C62a Have you experience any fever after touching sick or dead (died from illness) poultry? 1 Yes 0 No **If NO, Go to D1**
 តើអ្នកធ្លាប់ កើតគ្រុនក្តៅ ដែរឬទេ ក្រោយពីបានប៉ះពាល់ មាន់ទា ដែលឈឺ ឬ ងាប់ដោយសារជំងឺ ?

C62b Did you seek medical treatment for your symptoms? 1 Yes 0 No
 បើមាន តើ នៅពេលនោះ អ្នកបានស្វែងរកការព្យាបាលនៅទីណា ដែរឬទេ ?

D. QUESTIONS ON WILD BIRD MIXING (For head of the household only) ការលាយឡំគ្នារវាង មាន់ទា និង សត្វស្លាបព្រៃ (សំរាប់តែមេគ្រួសារ)

Next, I'd like to ask you a few questions about wild bird mixing with your flocks.

ខ្ញុំសូមសួររដ្ឋទាក់ទងនឹង ការលាយឡំគ្នារវាង មាន់ទារបស់អ្នក និង សត្វស្លាបព្រៃ

D1 Do your poultry mix with wild birds? 1 Everyday 2 Sometimes 0 Never **If NEVER, Go to D2**
 តើមាន់ទារបស់អ្នក ប៉ះពាល់ជាមួយសត្វស្លាបព្រៃរាល់ថ្ងៃដែរឬទេ? រាល់ថ្ងៃ ម្តងម្កាល មិនដែលសោះ

D1a What kind of wild birds? តើសត្វស្លាបព្រៃនោះជាសត្វអ្វី? _____

D1b Where do they mix? a Lake b Rice field c Farm
 តើមាន់ទារបស់អ្នក ទៅប៉ះពាល់ជាមួយសត្វស្លាបព្រៃនោះ នៅឯណា? បឹង វាលស្រែ ក្នុងផ្ទះ ឬ កសិដ្ឋាន

D2 Have you noticed any dead wild birds around your poultry areas? 1 Yes 0 No
 តើអ្នកមានដៃឃើញសត្វស្លាបព្រៃ ងាប់នៅក្បែររដ្ឋរបស់អ្នកដែរឬទេ?

D3 Do other animals (e.g. dogs, cats) carry dead wild birds onto your property? 1 Yes 0 No 9 Don't know
 តើមាន ឆ្កែ ឆ្កា ពាំ សត្វស្លាបព្រៃដែលងាប់ ចូលមកក្នុងបរិវេណផ្ទះរបស់អ្នកដែរឬទេ?

E. QUESTIONS ON ECONOMIC IMPACT OF BIRD FLU (For head of household only) ផលវិបាករបស់ជំងឺគ្រុនផ្តាសាយបក្សីលើជីវភាពគ្រួសារ (សំរាប់តែមេគ្រួសារ)

Lastly, I'd like to ask you a few questions about income from poultry. ចុងក្រោយនេះ ខ្ញុំសូមសួរអំពីប្រាក់ចំណូលរបស់អ្នក ដែលបានមកពីការចិញ្ចឹមមាន់ទា

E1 Is your income mainly from poultry raising? 1 Yes 0 No
 តើ ប្រាក់ចំណូល របស់គ្រួសារអ្នក ភាគច្រើនបានមកពីការចិញ្ចឹមមាន់ទា មែនឬទេ ?

E2 How much income do you generate per year from the sale of poultry? _____ 99 Don't know **If NO or Don't know, skip to E2b**
 តើក្នុង១ឆ្នាំ អ្នកអាចរកប្រាក់ចំណូលបានប៉ុន្មាន ពីការលក់ មាន់ទា របស់អ្នក? Add currency (Reil, Bath, Dollar) មិនដឹង

E2a What do you mainly spend it on? តើអ្នកចំណាយ ប្រាក់ដែលបានមក ពីការលក់ មាន់ទា របស់អ្នក ទៅលើអ្វីខ្លះ?
 a New poultry b School fee c Medical fee d Repaying loan e Pagoda f Clothes g Food
 ថ្លៃទិញមាន់ទាថ្មីយកមកចិញ្ចឹម ថ្លៃការរៀនសូត្រ ថ្លៃព្យាបាលជំងឺ សងថ្លៃជំពាក់គេ ទៅវត្ត ថ្លៃសំលៀកបំពាក់ ថ្លៃម្ហូបអាហារ
 h Other (Specify) ផ្សេងទៀត _____

E2b If you stopped selling poultry, how would it affect your household economy? បើសិនជាអ្នកឈប់ចិញ្ចឹម មាន់ទា តើអ្នកនឹងបាត់ប្រាក់ចំណូលច្រើនកម្រិតណា?
 1 No problem 2 Decrease half of income 3 Decrease most of income 4 Lose all income 9 Don't know
 គ្មានបញ្ហា បាត់ប្រាក់ចំណូលពាក់កណ្តាល បាត់ប្រាក់ចំណូល ១ភាគធំ បាត់ប្រាក់ចំណូលទាំងស្រុង មិនដឹង

E3 Have you ever vaccinated your poultry against AI? 1 Yes 0 No 9 DK **If NO or Don't know, go to Observation section**
 តើអ្នកធ្លាប់បានចាក់ វ៉ាក់សាំង ការពារមាន់ទា របស់អ្នក ប្រឆាំងនឹងជំងឺផ្តាសាយបក្សី ដែរឬទេ ?

E3a If yes, which flocks? 1 Chicken flocks only 2 Duck Flocks only 3 Both Chicken and duck flocks
 ចាក់បែបណា ? ចាក់ឱ្យតែមាន់ ចាក់ឱ្យតែទា ចាក់ឱ្យទាំងមាន់ ទាំងទា

I'd like to thank you for taking the time to answer my questions. Your involvement in this study is very important. Please accept this token of our appreciation as a thank you. (Hand out compensation kit)

ខ្ញុំបានបញ្ចប់សំណួរទាំងអស់ហើយ ដូចនេះ ខ្ញុំសូមអរគុណ ដែលអ្នកបានចំណាយពេលវេលា ឆ្លើយសំណួរទាំងអស់នេះ ។
 ការចូលរួមផ្តល់ចំណេះដឹងរបស់អ្នក ពិតជាមានសារៈសំខាន់ណាស់ ។ ខ្ញុំសូមជូនកញ្ចប់នេះ ទុកជាវត្ថុអនុស្សាវរីយ៍ ។

OBSERVATION Interviewer: Please take a few moments and fill in the following observational survey.
ការសង្កេត សូមបំពេញចំណើយខាងក្រោម ដោយធ្វើការសង្កេតនៅក្នុងបរិវេណផ្ទះ

01	See any DUCKS in the property	ឃើញមាន ទា នៅក្នុងទីធ្លា	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	If NO, Go to Q# 05	
02	Ducks in pond	ឃើញមាន ទា នៅក្នុងត្រពាំងទឹក	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> Don't have pond	គ្មានត្រពាំងទឹក
03	Ducks are free ranging	ឃើញមាន ទា ដើរដោយសេរី ក្នុងផ្ទះ	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
04	Ducks have contact pigs	ឃើញមាន ទា ទៅនៅលាយឡំជាមួយជ្រូក	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
05	See any CHICKENS in the property	ឃើញមាន មាន់ នៅក្នុងទីធ្លា	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	If NO, Go to Q# 08	
06	Chickens are free ranging	ឃើញមាន មាន់ ដើរដោយសេរី ក្នុងផ្ទះ	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
07	Chickens have contact with pigs	ឃើញមាន មាន់ ទៅនៅលាយឡំជាមួយជ្រូក	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
08	See poultry scrap (feathers...) on the property	ឃើញ មានសំណល់ពីធ្វើមាន់ទា នៅក្នុងទីធ្លា (ស្លាប...)	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
09	See poultry faeces on the property	ឃើញលាមកមាន់ទា រាយប៉ាយ នៅក្នុងទីធ្លា	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
010	See any protective material (used or not used) in the property, as listed:	ឃើញមានសម្ភារៈពាក់ការពារ (ទោះប្រើ ឬ មិនប្រើ) នៅក្នុងផ្ទះ ដូចជា:				
	a <input type="checkbox"/> Gloves ស្រោមដៃ	b <input type="checkbox"/> Rubber boots ស្បែកជើងដីរ ឃ្នូប ឬ ក វែង	c <input type="checkbox"/> Masks ម៉ាស់	d <input type="checkbox"/> Apron អាវការពារ	e <input type="checkbox"/> None មិនឃើញមានអ្វីទាំងអស់	
011	See anyone wearing protective material while caring for or preparing poultry for food	ឃើញអ្នកកំពុងសំអាតទ្រូងមាន់ទា ហើយមានពាក់ សម្ភារៈពាក់ការពារ	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		

F. Dengue Questions (For head of household only) **សំណួរ ទាក់ទងនឹងការអន្តរាគមន៍ លើជំងឺគ្រុនឈាម**

Have you received any of these materials or activities for dengue control in your household or your village ?
 តើអ្នកបានទទួលសម្ភារៈ ឬ សកម្មភាពអប់រំ ដូចរៀបរាប់ខាងក្រោមនេះ ដែរឬទេ នៅក្នុងភូមិ ឬ ក្នុងគ្រួសារ របស់អ្នក ?

Materials of activities សម្ភារៈ ឬ សកម្មភាព	[a] This year [2007] បានទទួល ក្នុងឆ្នាំ ២០០៧			[b] Previous years [before 2007] បានទទួល មុនឆ្នាំ ២០០៧		
	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK
F1 Impregnated bednet មុងជ្រលក់ថ្នាំ	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK
F2 Abate (larvicide agent) ថ្នាំអាបេត	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK
F3 Jar's lid (normal or impregnated) គំរូបពាង (មាន ឬ គ្មាន ជ្រលក់ថ្នាំ)	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK
F4 Spraying in the household ការចុះបាញ់ថ្នាំមូស	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK
F5 Dengue education session ការចុះអប់រំ តាមផ្ទះ ឬ ភូមិ	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK
F6 Dengue posters or brochures ផ្ទាំងរូបភាព ឬ ក្រដាសព័ត៌មាន អប់រំ	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> DK

SECTION C. QUESTIONS TO HOUSEHOLD MEMBERS សំណួរសំរាប់ សមាជិកគ្រួសារ ទូទៅ

Poultry mortality reporting ការផ្តល់ដំណឹងអំពី សត្វមាន់ទាមាប

I'm now going to ask you some questions about poultry mortality. ខ្ញុំសូមសួរបន្តទាក់ទងនឹងការផ្តល់ដំណឹងអំពី មាន់ទាមាប

C1 Have you experience poultry mortality in the since the Khmer New year? 1 Yes 0 No If NO, Go to Q# C7
 តើ មាន់ទា របស់អ្នក មានងាប់ខ្លះទេ តិចចាប់ពីពេលបុណ្យចូលឆ្នាំខ្មែរមក ?

What is the **total number of birds raised** since the Khmer New Year? (the subject may need some time to think about their answer)

C2T តើមាន **ចំនួនមាន់ សរុប** ដែលបានចិញ្ចឹម ទាំងអស់ប៉ុន្មានក្បាល ចាប់ពីបុណ្យចូលឆ្នាំខ្មែរមក? a Chickens ចំនួនមាន់ សរុប | | |

C3T តើមាន **ចំនួនទា សរុប** ដែលបានចិញ្ចឹម ទាំងអស់ប៉ុន្មានក្បាល ចាប់ពីបុណ្យចូលឆ្នាំខ្មែរមក? b Ducks ចំនួនទា សរុប | | |

C4T តើមាន **ចំនួនមេមាន់ សរុប** ដែលបានចិញ្ចឹម ទាំងអស់ប៉ុន្មានក្បាល ចាប់ពីបុណ្យចូលឆ្នាំខ្មែរមក? c Hens ចំនួនមេមាន់ សរុប | | |

Of those, how many of each species were **sick from illness** since the Khmer New Year?

C2S តើមាន **មាន់ ដែលឈឺ** សរុបទាំងអស់ប៉ុន្មានក្បាល? a Chickens ចំនួនមាន់ ឈឺ | | |

C3S តើមាន **ទា ដែលឈឺ** សរុបទាំងអស់ប៉ុន្មានក្បាល? b Ducks ចំនួនទា ឈឺ | | |

C4S តើមាន **មេមាន់ ដែលឈឺ** សរុបអស់ប៉ុន្មានក្បាល? c Hens ចំនួនមេមាន់ ឈឺ | | |

Of those that were sick from illness, how **many died from illness** since the Khmer New Year?

C2 តើមាន **មាន់ងាប់ ដោយជំងឺ** អស់ប៉ុន្មានក្បាល? a Chickens ចំនួនមាន់ងាប់ | | |

C3 តើមាន **ទាងាប់ ដោយជំងឺ** អស់ប៉ុន្មានក្បាល? b Ducks ចំនួនទាងាប់ | | |

C4 តើមាន **មេមាន់ងាប់ ដោយជំងឺ** អស់ប៉ុន្មានក្បាល? c Hens ចំនួនមេមាន់ងាប់ | | |

C5 Did you report poultry's mortality ? 1 Yes 0 No If NO, Go to Q# C7
 តើ អ្នកបានរាយការណ៍ប្រាប់អ្នកណា ដែរឬទេ អំពី មាន់ទា របស់អ្នក ដែលងាប់ ?

C6 To whom do you report? តើ អ្នកបានរាយការណ៍ប្រាប់ទៅអ្នកណាខ្លះ? (ចំណើយអាចមានច្រើន. កុំអានចំណើយ)

a Village chief ប្រធានភូមិ d District vet staff ពេទ្យសត្វស្រុក

b Village health staff / Health center ពេទ្យភូមិ ឬ នៅមណ្ឌលសុខភាព e Provincial vet staff ពេទ្យសត្វខេត្ត

c Village animal health worker / Vet staff ពេទ្យសត្វស្រុកត្រចិត្តតាមភូមិ ឬ ពេទ្យសត្វភូមិ f Other (specify) ផ្សេងទៀត _____

C7 If there is poultry mortality, what would make you report poultry mortality? (ចំណើយអាចមានច្រើន. កុំអានចំណើយ)

នៅពេលមានមាន់ ទា ងាប់ តើ មានអ្វីខ្លះមិនធម្មតា ដែលធ្វើឱ្យអ្នកទៅរាយការណ៍អំពី មាន់ទា របស់អ្នក ដែលងាប់ ?

- | | |
|--|--|
| a <input type="checkbox"/> Sudden death
មាន់ទា ងាប់រហ័សពេក

b <input type="checkbox"/> Half of flock die
មាន់ទា ងាប់អស់ពាក់កណ្តាលហ្នឹង | c <input type="checkbox"/> Whole flock die
មាន់ទា ទាំងហ្នឹងងាប់អស់រលីង

d <input type="checkbox"/> Other: _____
ផ្សេងទៀត |
|--|--|

C8 If there is a wild bird death, would you report it? 1 Yes 0 No

ប្រសិនបើអ្នកជួបប្រទះ សត្វស្លាប់ព្រៃដែលងាប់ តើអ្នកនឹងទៅរាយការណ៍ ដែរឬទេ ?

Practices when mortality occurred ការអនុវត្តនៅពេលមាន ម៉ាន់ទាងាប

C9 If you encounter poultry mortality in your flocks, what do you do with dead poultry?

នៅពេលដែលមាន ម៉ាន់ទាងាបក្នុងផ្ទះរបស់អ្នក តើអ្នកយក ម៉ាន់ទា របស់អ្នក ដែលងាប់ ទៅធ្វើអ្វីខ្លះ?

(សូមអានចំលើយ)

- [a] Bin បោះចោលនៅធុងសំរាម 1 Yes 0 No
- [b] Bury កប់ចោល 1 Yes 0 No If no, skip to d
- [c] Can you point out where you buried them? [Check **Yes** only when they can show you where they buried poultry] 1 Yes 0 No
 សូមបង្ហាញកន្លែងដែលអ្នកបានកប់ [គូស YES នៅពេលណាដែលគេអាចបង្ហាញកន្លែងកប់]
- [d] Burn ដុតចោល 1 Yes 0 No If no, skip to f
- [e] Can you point out where you burned them? [Check **Yes** only when they can show you where they burned poultry] 1 Yes 0 No
 សូមបង្ហាញកន្លែងដែលអ្នកបានដុត [គូស YES នៅពេលណាដែលគេអាចបង្ហាញកន្លែងដុត]
- [f] Feed other animals បោះអោយសត្វផ្សេងស្តី 1 Yes 0 No
- [g] Prepare for selling ធ្វើលក់ 1 Yes 0 No If no, skip to i
- [h] Where did you sell? តើអ្នក យកទៅលក់ នៅឯណា? (Village, Commune, District, Province) _____
- [i] Prepare for food យកមកចម្អិនធ្វើម្ហូប 1 Yes 0 No
- [j] Sell carcass លក់ម៉ាន់ទាងាបទាំងមូល 1 Yes 0 No If no, skip to l
- [k] Where did you sell? តើអ្នក យកទៅលក់ នៅឯណា? (Village, Commune, District, Province) _____
- [l] Give away to neighbor ឱ្យទៅអ្នកជិតខាង 1 Yes 0 No
- [m] Throw into water sources បោះចោលទៅក្នុងទឹក 1 Yes 0 No
- [n] Throw away ត្រូវវែងចោល 1 Yes 0 No
- [o] Other (specify) ផ្សេងទៀត _____

C10 What do you do if/when you have sick poultry in your flocks ?

នៅពេលដែលមាន ម៉ាន់ទាលី ក្នុងផ្ទះរបស់អ្នក តើអ្នកយក ម៉ាន់ទា របស់អ្នក ដែលលី ទៅធ្វើអ្វីខ្លះ?

(សូមអានចំលើយ)

Would you:

- [a] Quarantine separately from other in the flocks? ដាក់ ម៉ាន់ទា ដែលលី ដាច់ដោយឡែកពី ម៉ាន់ទាដទៃទៀត 1 Yes 0 No
- [b] Slaughter for selling? សំលាប់យកសាច់លក់ 1 Yes 0 No If no, skip to e
- [c] Where did you sell? តើអ្នកយកទៅលក់នៅឯណា? (village, commune, district, Province) _____
- [d] Did you use a middleman? តើអ្នកមានលក់ទៅឱ្យឈ្មួញដើរទិញម៉ាន់ទាដែរឬទេ? 1 Yes 0 No
- [e] Slaughter for food? សំលាប់យកសាច់ធ្វើម្ហូប 1 Yes 0 No
- [f] Give to neighbors for consumption? ឱ្យទៅអ្នកជិតខាងយកទៅធ្វើម្ហូប 1 Yes 0 No
- [g] Sell alive? លក់ទាំងរស់ 1 Yes 0 No If no, skip to j
- [h] Where did you sell? តើអ្នកយកទៅលក់នៅឯណា? (village, commune, district, Province) _____
- [i] Did you use a middleman? តើអ្នកមានលក់ទៅឱ្យឈ្មួញដើរទិញម៉ាន់ទាដែរឬទេ? 1 Yes 0 No
- [j] Do nothing? មិនបានធ្វើអ្វីទាំងអស់ 1 Yes 0 No
- [k] Other (specify) ផ្សេងទៀត _____

C11 What do you do with remaining poultry that survived in the flock ?

តើអ្នកយក មាន់ទា របស់អ្នក ដែលនៅសល់ពីលើ ឬ ងាប់ ទៅធ្វើអ្វីខ្លះ?

(សូមអានចំណើយ)

Would you:

- [a] Quarantine separately from other in the flocks? ដាក់ មាន់ទា ដែលលើ ដាច់ដោយឡែកពី មាន់ទាដទៃទៀត 1 Yes 0 No
- [b] Slaughter for selling? សំលាប់យកសាច់លក់ 1 Yes 0 No If no, skip to e
- [c] Where did you sell? តើអ្នកយកទៅលក់នៅណា? (village, commune, district, Province) _____
- [d] Did you use a middleman? តើអ្នកមានលក់ទៅឱ្យល្អញូដើរទិញមាន់ទាដែរឬទេ? 1 Yes 0 No
- [e] Slaughter for food? សំលាប់យកសាច់ធ្វើម្ហូប 1 Yes 0 No
- [f] Give to neighbors for consumption? ឱ្យទៅអ្នកជិតខាងយកទៅធ្វើម្ហូប 1 Yes 0 No
- [g] Sell alive? លក់ទាំងរស់ 1 Yes 0 No If no, skip to j
- [h] Where did you sell? តើអ្នកយកទៅលក់នៅណា? (village, commune, district, Province) _____
- [i] Did you use a middleman? តើអ្នកមានលក់ទៅឱ្យល្អញូដើរទិញមាន់ទាដែរឬទេ? 1 Yes 0 No
- [j] Do nothing? មិនបានធ្វើអ្វីទាំងអស់ 1 Yes 0 No
- [k] Other (specify) ផ្សេងទៀត _____

Knowledge and attitude ចំណេះដឹង និង ការយល់

- C12 Do you think it's important to report poultry mortality? 1 Yes If YES, Go to Q# C12b
- តើអ្នកគិតថាការរាយការណ៍ អំពី មាន់ទា ងាប់ មានសារៈសំខាន់ដែរឬទេ? 0 No If NO, Go to Q# C12a
- 9 Don't know If DK, Go to Q# C13

C12a **If NO,** Why is it not important? ហេតុអ្វីបានជាអ្នកគិតថាការរាយការណ៍ អំពី មាន់ទា ងាប់ **មិនមាន** សារៈសំខាន់? (ចំណើយអាចមានច្រើន, កុំអានចំណើយ)

- a No help from vet staff or authorities ព្រោះមិនមានជំនួយពីពេទ្យសត្វ ឬ អាជ្ញាធរ d Don't know មិនដឹង
- b Too few death ព្រោះមាន់ទាងាប់តិចតួច e Other (specify) ផ្សេងទៀត
- c Poultry are not important (like pigs or cattles..) ព្រោះមាន់ទាមិនសំខាន់ដូចសត្វគោក្របីទេ

C12b **If YES,** Why is it important? ហេតុអ្វីបានជាអ្នកគិតថាការរាយការណ៍ អំពី មាន់ទា ងាប់ **មាន សារៈសំខាន់**? (ចំណើយអាចមានច្រើន, កុំអានចំណើយ)

- a It could be AI ព្រោះអាចមានជំងឺគ្រុនផ្តាសាយបក្សី c Don't know មិនដឹង
- b We can have advices from VAHWs ព្រោះគិតថាអាចទទួលបានការណែនាំពីបុគ្គលិកពេទ្យសត្វ d Other (specify) ផ្សេងទៀត

C13 If you report mortality, what do you expect in return? បើអ្នករាយការណ៍ តើអ្នកចង់បានអ្វីខ្លះត្រលប់មកវិញ?

C13a And from whom? តើអ្នកចង់បានពីអ្នកណា? _____

C14 What do you think discourages villagers to report poultry mortality?

តាមយោបល់របស់អ្នក តើអ្វីខ្លះដែលធ្វើឱ្យអ្នកភូមិមិនចង់ទៅរាយការណ៍ អំពីមាន់ទាដែលងាប់?

(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)

- a Fear of poultry culling ខ្លាចគេមកសំលាប់មាន់ទារបស់ពួកគេអស់ c Fear of panic in the village ខ្លាចធ្វើឱ្យមានការភ័យច្រៀតច្របល់នៅក្នុងភូមិ
- b Problems with selling ខ្លាចលក់មាន់ទាលែងបាន d Don't know មិនដឹង
- e Don't know where/to whom to report មិនសឹងទៅប្រាប់អ្នកណា, នៅទីណា
- f Poultry are not important like cattle មាន់ទាជាសត្វតូចតាច មិនសំខាន់ដូចគោ ក្របីទេ
- g Other? (specify) ផ្សេងទៀត _____

C15 What, do you think, would help encourage farmers to report poultry's mortality ?

តាមយោបល់របស់អ្នក តើអ្វីខ្លះដែលអាចជួយលើកទឹកចិត្តអ្នកភូមិឱ្យ ទៅរាយការណ៍ អំពីមាន់ទាដែលងាប់?

(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)

- a Awareness / education on the risk of AI ការអប់រំអំពីគ្រោះថ្នាក់បណ្តាលមកពីជំងឺគ្រុនផ្តាសាយបក្សី d Incentive ការផ្តល់ជំរាញដល់អ្នកទៅរាយការណ៍
- b Help from authorities (drug, food, training...) ជំនួយពីអាជ្ញាធរ (ឧ: ថ្នាំ, ចំណីសត្វ, វគ្គបណ្តុះបណ្តាល...) e Buying sick or dead poultry at lower price ជួយទិញមាន់ទាដែល លើ ឬ ងាប់ ដោយតម្លៃថោកៗ
- c Compensation in case of culling មានជាសំណងក្នុងករណីសំលាប់មាន់ទា f Don't know មិនដឹង
- g Ask them to report ស្នើរសុំឱ្យគាត់ទៅរាយការណ៍
- h Other (specify) ផ្សេងទៀត _____

28	Do you cook or help in cooking poultry for your family ? តើអ្នក ធ្វើម្ហូប ឬ ជួយធ្វើម្ហូប ដែរឬទេ?	1 <input type="checkbox"/> Everyday រាល់ថ្ងៃ	2 <input type="checkbox"/> Sometimes ម្តងម្កាល	0 <input type="checkbox"/> Never មិនដែលសោះ
29	Do you: Boil birds? តើអ្នក ស្ទោរចំអិនមាន់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
9a	Slaughter/ bleed birds? តើអ្នក អារក សំលាប់ មាន់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
9b	Cut meat? តើអ្នក កាប់,ហាន់ សាច់មាន់ សាច់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
9c	Wash meat? តើអ្នក លាង សាច់មាន់ សាច់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
9d	Remove internal organs from birds? តើអ្នក រះយកគ្រឿងក្នុង មាន់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
9e	Wash internal organs? តើអ្នក លាង គ្រឿងក្នុង មាន់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
f	Do you eat raw or half cooked chicken eggs (eggs with runny yokes)? តើអ្នក ហូប ឬ ហុត ពងមាន់នៅ ឬ ចំអិន នៅដីរញ្ឆៅ ដែរឬទេ ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
9g	Do you eat raw chicken meat? តើអ្នក ហូប ម្ហូបដែលធ្វើពី សាច់មាន់ នៅ ដែរឬទេ ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
h	Do you eat chicken meat that is pink in color (has "pink spots")? តើអ្នក ហូប សាច់មាន់ ដែលចំអិន នៅនៅ ក្រហមដុំៗ ដែរឬទេ ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
i	Do you eat raw or half cooked duck eggs (eggs with runny yokes)? តើអ្នក ហូប ឬ ហុត ពងទា នៅ ឬ ចំអិន នៅដីរញ្ឆៅ ដែរឬទេ ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
j	Do you eat raw duck meat? តើអ្នក ហូប ម្ហូបដែលធ្វើពីសាច់ទា នៅ ដែរឬទេ ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
k	Do you eat duck meat that is pink in color (has "pink spots")? តើអ្នក ហូប សាច់ទា ដែលចំអិន នៅនៅ ក្រហមដុំៗ ដែរឬទេ ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
30	Have you ever <u>prepared</u> wild birds for food? តើអ្នកធ្លាប់ ធ្វើ,កាប់ សាច់សត្វស្លាបព្រៃចំអិនធ្វើម្ហូប ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
31	Do you prepare poultry near pond, river, water well? តើអ្នក មានយកមាន់ទៅធ្វើ នៅក្បែរ ត្រពាំង ព្រែក ទន្លេ ឬ អណ្តូង ដែរឬទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> No pond/rever/ open water well មិនមាន ត្រពាំង ព្រែក ទន្លេ ឬ អណ្តូង
32	Do wash poultry products directly in the water source (pond/river)? តើអ្នក មានយកមាន់ទៅ លាងផ្ទាល់ នៅក្នុង ត្រពាំង ព្រែក ឬ ទន្លេ ដែរឬទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> No pond/rever មិនមាន ត្រពាំង ព្រែក ឬ ទន្លេ
33	Do you care or help care for poultry? តើអ្នក មានជួយមើលថែទាំ មាន់ទា ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No If NO, go to Q35
3a	How many times a week do you clean your poultry cages/areas ? តើអ្នក សំអាត ទ្រុងមាន់ទា ប៉ុន្មានដងក្នុង១អាទិត្យ?			times per week 0 <input type="checkbox"/> <1 time a week
34	When caring for poultry: នៅពេលដែលអ្នក ថែទាំ សំអាត ទ្រុងមាន់ទា			
4a	Do you wear plastic bags over hands? តើអ្នក មានពាក់ថង់ផ្លាស្ទិក ការពារនៅដៃ ដែរឬទេ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No
4b	Do you have any gloves in your household? តើអ្នក មានស្រោមដៃដែរ ដែរឬទេ នៅក្នុងផ្ទះរបស់អ្នក?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No If NO, Go to C34d
4c	How often do wear gloves when touching poultry? តើអ្នក ពាក់ស្រោមដៃ រាល់ដងដែរឬទេ នៅពេលប៉ះពាល់មាន់ទា?	1 <input type="checkbox"/> Everyday	2 <input type="checkbox"/> Sometimes	0 <input type="checkbox"/> Never មិនដែលពាក់សោះ
4d	Do you have any rubber boots in your household? តើអ្នក មានស្បែកជើងជ័រ ក វែង ដែរឬទេ នៅក្នុងផ្ទះ?			1 <input type="checkbox"/> Yes 0 <input type="checkbox"/> No If NO, Go to C34f
4e	How often do you wear rubber boots when caring for poultry? តើអ្នក ពាក់ស្បែកជើងជ័រ ក វែង រាល់ដងដែរឬទេ នៅពេលធ្វើការសំអាតទ្រុងមាន់ទា?	1 <input type="checkbox"/> Everyday	2 <input type="checkbox"/> Sometimes	0 <input type="checkbox"/> Never មិនដែលពាក់សោះ

4f Do you have any aprons in your household? តើអ្នក មានអាវកាតារ ដែរឬទេ នៅក្នុងផ្ទះរបស់អ្នក? 1 [] Yes 0 [] No **If NO, Go to C34h**
 4g How often do you wear an apron when caring for poultry? 1 [] Everyday 2 [] Sometimes 0 [] Never
 តើអ្នក ពាក់អាវកាតារ រាល់ដងដែរឬទេ នៅពេលប៉ះពាល់ ថែទាំ មាន់ទា? ពាក់រាល់ដង ពាក់ម្តងម្កាល មិនដែលពាក់សោះ

4h Do you have any masks in you household? តើអ្នក មានម៉ាស់ (របាំងមុខ) ដែរឬទេ នៅក្នុងផ្ទះរបស់អ្នក? 1 [] Yes 0 [] No **If NO, Go to C35**
 4i How often do you wear face mask when caring for poultry? 1 [] Everyday 2 [] Sometimes 0 [] Never
 តើអ្នក ពាក់ម៉ាស់ (របាំងមុខ) ការពារ រាល់ដងដែរឬទេ នៅពេលប៉ះពាល់ ថែទាំ មាន់ទា? ពាក់រាល់ដង ពាក់ម្តងម្កាល មិនដែលពាក់សោះ

35 Do you, personally, feed your poultry ? តើអ្នក ឱ្យចំណី មាន់ទា ដោយខ្លួនឯង ឬទេ? 1 [] Yes 0 [] No **If NO, Go to C36**
 5a How many times a week do you, personally, feed poultry? តើអ្នក ឱ្យចំណី មាន់ទា ប៉ុន្មានដង ក្នុង១អាទិត្យ? | | | Times 0 [] <1 time a week
 36 Do your poultry go in rice fields or lake ? តើមាន់ទារបស់អ្នកចេញដើរកសិ តាមវាលស្រែ ឬ បឹង ដែរឬទេ? 1 [] Yes 0 [] No
 37 Do you use poultry faeces for manure ? តើអ្នកប្រើលាមកមាន់ទា សំរាប់ធ្វើដី ដែរឬទេ? 1 [] Yes 0 [] No

38 Have you ever been to Vietnam ? តើអ្នក ធ្លាប់ទៅដល់ប្រទេសវៀតណាម ដែរឬទេ? 1 [] Yes 0 [] No **If NO, Go to C39**
 3a How many times per month do you go to Vietman? តើអ្នក ទៅប៉ុន្មានដង ក្នុង១ខែ? | | | times per month 0 [] <1 time a month
 3b Do you go there for trade (sell or buy) poultry or poultry products? អ្នក ទៅដើម្បីលក់ , ទិញ មាន់ទា តើមែនឬទេ? 1 [] Yes 0 [] No **If no, go to C39**
 3c Which provinces do you go to sell/buy? អ្នក ទៅដល់ខេត្តណាខ្លះ? _____

39 Have you ever been to Thailand ? តើអ្នក ធ្លាប់ទៅដល់ប្រទេសថៃ ដែរឬទេ? 1 [] Yes 0 [] No **If NO, Go to C40s**
 3a How many times per month do you go to Thailand? តើអ្នក ទៅប៉ុន្មានដង ក្នុង១ខែ? | | | times per month 0 [] <1 time a month
 3b Do you go there for trade (sell or buy) poultry or poultry products? អ្នក ទៅដើម្បីលក់ , ទិញ មាន់ទា តើមែនឬទេ? 1 [] Yes 0 [] No **If no, go to C40s**
 3c Which provinces do you go to sell/buy? អ្នក ទៅដល់ខេត្តណាខ្លះ? _____

Hygiene practices ការអនុវត្ត អនាម័យជាមូលដ្ឋាន
 40s Do you have soap in your household? តើ អ្នកមានសាប៊ូ ឬទេ នៅក្នុងផ្ទះរបស់អ្នក ? 1 [] Yes 0 [] No
 40 Do you wash your hands with soap after you touch poultry? 1 [] Yes 0 [] No **If NO, Go to C40b**
 តើអ្នក លាងដៃជាមួយសាប៊ូ ដែរឬទេ ក្រោយពេលប៉ះពាល់មាន់ទា?
 4a How often? តើអ្នក លាង ជាមួយសាប៊ូ រាល់ដងដែរឬទេ? 1 [] Everytime 2 [] Sometimes 3 [] Rarely
 4b If you have no soap or run out of soap, what do you use instead of soap to wash your hand?
 បើអ្នកគ្មានសាប៊ូ តើអ្នក ប្រើអ្វីជំនួស សំរាប់លាងដៃ? _____
 41 Which water source do you usually use in your household? តើអ្នក យកទឹកអ្វី មកពីណា សំរាប់ប្រើប្រាស់ក្នុងគ្រួសារ? **(ចំណើយអាចមានច្រើន)**
 a [] Open water well ទឹកអណ្តូងយោង c [] Ponds ទឹកត្រពាំង, ស្រះ e [] Lake ទឹកបឹង g [] Other (specify) _____
 b [] Water well with pump ទឹកអណ្តូងស្តាប់ d [] Water tap ទឹកម៉ាស៊ីន f [] River ទឹកទន្លេ

C42 If pond or lake or river does, poultry has access to those water sources as well? 1 Yes 0 No 9 Don't Know
 បើ ត្រពាំង បឹង ឬ ទន្លេ តើ មាន ម៉ាន់ ចុះ ទៅ ក្នុង ទឹក នោះ ដែរ ឬ ទេ?

C42a Which water source do you used to bathe? តើ អ្នក ងូត ទឹក អ្វី នៅ ឯណា ? (ចំលើយ អាច មាន ច្រើន)
 a Open water well ទឹក អណ្តូង យោង
 c Ponds ទឹក ត្រពាំង, ស្រះ
 e Lake ទឹក បឹង
 g Other (specify) _____ ផ្សេង ទៀត
 b Water well with pump ទឹក អណ្តូង ស្នប់
 d Water tap ទឹក ម៉ាស៊ីន
 f River ទឹក ទន្លេ

C42b If pond or lake or river, do poultry have access to these water sources? 1 Yes 0 No 9 Don't Know
 បើ ត្រពាំង បឹង ឬ ទន្លេ តើ មាន ម៉ាន់ ចុះ ទៅ ក្នុង ទឹក នោះ ដែរ ឬ ទេ?

Knowledge about AI / bird flu ចំណេះដឹងអំពី ជំងឺ គ្រុនផ្តាសាយ បក្សី

I'm now going to ask you some questions about avian influenza. ខ្ញុំ សូម សួរ បន្ត ទាក់ ទង នឹង ជំងឺ គ្រុនផ្តាសាយ បក្សី

C43 Have you heard of AI / bird flu? តើ អ្នក ធ្លាប់ លឺ អំពី ជំងឺ គ្រុនផ្តាសាយ បក្សី ដែរ ឬ ទេ? 1 Yes 0 No **If NO, Go to C47**

C44 Where did you hear that information from? តើ អ្នក ធ្លាប់ លឺ អំពី ជំងឺ គ្រុនផ្តាសាយ បក្សី មក ពី ណា ខ្លះ ? (ចំលើយ អាច មាន ច្រើន, កុំ អាន ចំលើយ)
 a Village vet staffs ពេទ្យ សត្វ ភូមិ
 d Radio វិទ្យុ
 g Public poster ផ្ទាំង រូប ភាព អប់រំ តាម ភូមិ
 j Someone in the family សមាជិក គ្រួសារ
 b Health staff / health center ពេទ្យ ភូមិ ឬ នៅ មណ្ឌល សុខ ភាព
 e Television ទូរទស្សន៍
 h Brochures កូន សៀវភៅ អប់រំ វត្ត ច្បាញ
 k Other (specify) _____ ផ្សេង ទៀត
 c Village chiefs ប្រធាន ភូមិ
 f Newspaper កាសែត
 i NGO health education sessions វគ្គ អប់រំ របស់ អង្គការ នានា

C44i What would you consider to be your MAIN source of AI information? (open question) _____
 ក្នុង ចំណោម ប្រភព ទាំង នោះ តើ ប្រភព មួយ ណា ដែល អ្នក គិត ថា បាន ផ្តល់ ព័ត៌មាន អំពី ផ្តាសាយ បក្សី ដល់ អ្នក បាន ច្រើន ជាង គេ ?

C45 Have you taught your children about AI តើ អ្នក ធ្លាប់ បង្រៀន កូន ៗ អ្នក ឬ ក្មេង ៗ អំពី ជំងឺ គ្រុនផ្តាសាយ បក្សី ដែរ ឬ ទេ? 1 Yes 0 No **If NO, Go to C46**

C45a What have you taught them (open question) _____
 តើ អ្នក ប្រាប់ កូន ៗ អ្នក យ៉ាង ដូច ម្តេច ខ្លះ ?

C46 In your own words, what is bird flu ? តាម យោបល់ របស់ អ្នក តើ ជំងឺ គ្រុនផ្តាសាយ បក្សី ជា អ្វី ? (ចំលើយ អាច មាន ច្រើន, កុំ អាន ចំលើយ)
 a Poultry disease (duck/chicken...) ជំងឺ ម៉ាន់ ៗ
 c Human disease ជំងឺ មនុស្ស
 e Pig disease ជំងឺ ជ្រូក
 g Can be transmitted to human អាច ឆ្លង មក មនុស្ស បាន
 b Can kill poultry អាច សំលាប់ ម៉ាន់ ៗ បាន
 d Can kill human អាច សំលាប់ មនុស្ស បាន
 f Wild bird disease ជំងឺ សត្វ ល្អិត ប្រៃ
 h Other (specify) ផ្សេង ទៀត

C47 Could you tell if your flock were infected by AI? 1 Yes 0 No 9 Don't know
 តើ អ្នក គិត ថា ម៉ាន់ ៗ របស់ អ្នក ធ្លាប់ មាន ជំងឺ គ្រុនផ្តាសាយ បក្សី ពី មុន មក ដែរ ឬ ទេ?

C48 Do you think your poultry are at risk of being infected with AI? 1 Yes 0 No 9 Don't know
 តើ អ្នក គិត ថា ម៉ាន់ ៗ របស់ អ្នក អាច ឆ្លង ជំងឺ គ្រុនផ្តាសាយ បក្សី ដែរ ឬ ទេ?

C49 When chickens are sick, Do you know what are SYMPTOMS that make you suspect of AI infection in CHICKENS? 1 Yes
នៅពេលមាន ម្ចាស់ឈឺ តើ អ្នកស្គាល់រោគសញ្ញា ដែលធ្វើឱ្យអ្នកសង្ស័យថា ម្ចាស់ ទាំងនោះ មានជំងឺគ្រុនផ្តាសាយបក្សី ដែរឬទេ? 0 No **If NO, Go to C51**

C50 What are SYMPTOMS that make you suspect of AI infection in CHICKENS? 9 Don't know **If DK, Go to C51**

តើ រោគសញ្ញា អ្វីខ្លះដែលធ្វើឱ្យអ្នកសង្ស័យថា ម្ចាស់ ដែលឈឺទាំងនោះ មានជំងឺគ្រុនផ្តាសាយបក្សី?

a Sudden death (1-2 days)
ងាប់ឈឺរឿន " ១ - ២ ថ្ងៃ "

f Stop laying eggs
ឈប់ពង

(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)

k White / cloudy / blind eyes
ភ្នែកស

b Sudden death in large number
ងាប់ឈឺរឿនហើយក្នុងចំនួនច្រើនក្បាល

g Eat less / Stop eating
លែងស្ងូរ ឬ អត់ស៊ីចំណី

l Difficult/Fast Breathing
ពិបាកដកដង្ហើម

c Motionless / Sleepiness
សំកុក / ដេក

h Violet / swollen combs and wattles / head
ហើមជាំក្បាល និង សិរណង់

m Leg weakness
ទន់ជើង

d Seizure / Spining
ប្រកាច់ / ដើរវិលៗ

i Neck Twist
ទន់ក ឬ បត់ក

n Diarrhea
រាត

e Bleeding
ចេញឈាម

j Eye / nose discharge
ហៀរទឹកភ្នែក/ហៀរសំបោរ

o Ruffled feathers
ជ្រុះស្នាប ឬ រោម

p Other (Specify) _____

ផ្សេងទៀត

C51 When ducks are sick, Do you know what are SYMPTOMS that make you suspect of AI infection in DUCKS? 1 Yes
នៅពេលមាន ម្ចាស់ឈឺ តើ អ្នកស្គាល់រោគសញ្ញា ដែលធ្វើឱ្យអ្នកសង្ស័យថា ម្ចាស់ ទាំងនោះ មានជំងឺគ្រុនផ្តាសាយបក្សី ដែរឬទេ? 0 No **If NO, Go to C53**

C52 What are SYMPTOMS that make you suspect of AI infection in DUCKS? 9 Don't know **If DK, Go to C53**

តើ រោគសញ្ញា អ្វីខ្លះដែលធ្វើឱ្យអ្នកសង្ស័យថា ម្ចាស់ ដែលឈឺទាំងនោះ មានជំងឺគ្រុនផ្តាសាយបក្សី?

a Sudden death (1-2 days)
ងាប់ឈឺរឿន " ១ - ២ ថ្ងៃ "

f Stop laying eggs
ឈប់ពង

k White / cloudy / blind eyes
ភ្នែកស

b Sudden death in large number
ងាប់ឈឺរឿនហើយក្នុងចំនួនច្រើនក្បាល

g Eat less / Stop eating
លែងស្ងូរ ឬ អត់ស៊ីចំណី

l Difficult/Fast Breathing
ពិបាកដកដង្ហើម

c Motionless / Sleepiness
សំកុក / ដេក

h Violet / swollen combs and wattles / head
ហើមជាំក្បាល និង សិរណង់

m Leg weakness
ទន់ជើង

d Seizure / Spining
ប្រកាច់ / ដើរវិលៗ

i Neck Twist
ទន់ក ឬ បត់ក

n Diarrhea
រាត

e Bleeding
ចេញឈាម

j Eye / nose discharge
ហៀរទឹកភ្នែក/ហៀរសំបោរ

o Ruffled feathers
ជ្រុះស្នាប ឬ រោម

p Other (Specify) ផ្សេងទៀត _____

C53 How is AI spread among poultry / birds? តើជំងឺគ្រុនផ្តាសាយបក្សី ឆ្លងរាលដាលក្នុងចំណោម ម្ចាស់ ទា ដោយរបៀបណា? (ចំណើយអាចមានច្រើន, កុំអានចំណើយ)

a Contact with another infected/sick birds
ប៉ះពាល់សត្វមានទា គ្នាវា ដែលឈឺ?

d Contact with virus brought in by people, their clothing or footwear
មានមនុស្សមកពីក្រៅ ដោយមានជាប់មេរោគតាមខ្នងចូលមក?

b Contact with infected faeces?
ប៉ះពាល់ជាមួយឈាមកម្លាំងទា គ្នាវា ដែលឈឺ?

e Don't know
មិនដឹង

c Contact with other contaminated feed
ការប៉ះពាល់ ស៊ី ចំណីសត្វមិនស្អាតដែលមានផ្ទុកមេរោគ

f Other (Specify) _____
ផ្សេងទៀត

C54 Can humans get AI? តើមនុស្សអាចឆ្លងជំងឺគ្រុនផ្តាសាយបក្សីដែរឬទេ? 1 [] Yes 0 [] No 9 [] Don't know **If No or Don't know, skip to C56**

C55 How can humans catch AI? តើជំងឺគ្រុនផ្តាសាយបក្សីឆ្លងមកមនុស្ស ដោយរបៀបណា? **(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)**

- a [] Contact with sick/dead poultry ការប៉ះពាល់មាន់ទាប្លឺស្លាប់
- b [] Contact with poultry feces ការប៉ះពាល់អាចម៍សត្វ
- c [] Eating undercooked poultry products បរិភោគមាន់មិនទាន់ឆ្អិនល្អ " សាច់ ស៊ុត .."
- d [] Eating raw poultry products (eggs / blood pudding) បរិភោគឆ្មៅ " ស៊ុត សាច់ ឈាម .."
- e [] Contact with contaminated farm equipment ការប៉ះពាល់សំភារៈកសិដ្ឋានមានផ្ទុកមេរោគ
- f [] Don't know មិនដឹង
- g [] Other (Specify) ផ្សេងទៀត _____

C56 Do you think Bird flu can kill (is fatal for) humans? តើអ្នកគិតថា ជំងឺគ្រុនផ្តាសាយបក្សី អាចសំលាប់មនុស្សបាន ដែរឬទេ? 1 [] Yes 0 [] No 9 [] Don't know

C57 In Cambodia, Have there been any fatal human cases of AI? តើមានមនុស្សស្លាប់ដោយ ជំងឺគ្រុនផ្តាសាយបក្សី ដែរឬទេ នៅប្រទេសកម្ពុជាយើង? 1 [] Yes 0 [] No 9 [] Don't know

C58 Do you think AI can be transmitted to **HUMANS** by... តើអ្នកគិតថា ជំងឺគ្រុនផ្តាសាយបក្សីឆ្លងមកមនុស្ស តាម...

- [a] Eating well cooked poultry's products ? ហូបសាច់ ស៊ុត មាន់ទាដែលបានចម្អិនឆ្អិនល្អ? 1 [] Yes 0 [] No 9 [] Don't know
- [b] Eating undercooked poultry's products ? ហូបសាច់ ស៊ុត មាន់ទាដែល ចម្អិននៅឆ្នៅ មិនឆ្អិនល្អ? 1 [] Yes 0 [] No 9 [] Don't know
- [c] Eating poultry eggs from healthy chickens? ហូបពងមាន់ ពងទា? 1 [] Yes 0 [] No 9 [] Don't know
- [d] Eating poultry bought from market? ហូបសាច់មាន់ទា ដែលទិញយកមកពីផ្សារ? 1 [] Yes 0 [] No 9 [] Don't know
- [e] Eating your own poultry prepared at home? ហូបសាច់មាន់ទា ដែលចិញ្ចឹម និង ធ្វើ ដោយខ្លួនឯង? 1 [] Yes 0 [] No 9 [] Don't know
- [f] Eating poultry imported from outside Cambodia? ហូបសាច់មាន់ទា ដែលនាំចូលមកពីក្រៅប្រទេសកម្ពុជា? 1 [] Yes 0 [] No 9 [] Don't know
- [g] Swimming in ponds? មុជទឹកក្នុងស្រះ 1 [] Yes 0 [] No 9 [] Don't know
- [h] Touching wild birds ? ប៉ះពាល់សត្វស្លាបព្រៃ? 1 [] Yes 0 [] No 9 [] Don't know
- [i] Eating wild birds? ហូបសាច់សត្វស្លាបព្រៃ? 1 [] Yes 0 [] No 9 [] Don't know
- [j] From other people? ពីមនុស្សម្នាក់ ទៅ ម្នាក់ទៀត 1 [] Yes 0 [] No 9 [] Don't know
- [k] Touching healthy poultry ? ប៉ះពាល់សត្វមាន់ទា អត់មានជំងឺ? 1 [] Yes 0 [] No 9 [] Don't know
- [l] Touching sick or dead poultry with bare hands? ប៉ះពាល់សត្វមាន់ទា ដែលឈឺ ងាប់ ដោយដៃទទេ? 1 [] Yes 0 [] No 9 [] Don't know
- [m] Touching poultry faeces? ប៉ះពាល់ជាមួយឈាមសត្វមាន់ទា? 1 [] Yes 0 [] No 9 [] Don't know
- [n] From sexual contact? តាមការរួមភេទ 1 [] Yes 0 [] No 9 [] Don't know
- [o] Touching poultry blood? ប៉ះពាល់ជាមួយ ឈាមសត្វមាន់ទា? 1 [] Yes 0 [] No 9 [] Don't know
- [p] Slaughtering poultry ? សំលាប់មាន់ទាធ្វើម្ហូប? 1 [] Yes 0 [] No 9 [] Don't know
- [q] Using the same cutting board for poultry and food products? ប្រើជ្រុងក្រែក រួម សំរាប់ សាច់មាន់ ទា និង បន្លែ សាច់ គ្រប់មុខ ដទៃទៀត? 1 [] Yes 0 [] No 9 [] Don't know

C59 Do you think AI can be transmitted to CHILDREN by touching / playing with sick / dead poultry (with bare hands)? តើអ្នកគិតថា កូនក្មេង អាចឆ្លងជំងឺគ្រុនផ្តាសាយបក្សី ដោយ ប៉ះពាល់ ឬ លេង ជាមួយ មាន់ទាដែល ឈឺ ឬ ងាប់ ដែរឬទេ? 1 [] Yes 0 [] No 9 [] Don't know

C60 Do you know any AI / Bird Flu signs and symptoms in **HUMANS**? តើអ្នកស្គាល់រោគសញ្ញាជំងឺគ្រុនផ្តាសាយបក្សី នៅក្នុងខ្លួន **មនុស្ស** ដែរឬទេ? 1 [] Yes 0 [] No **If NO, Go to C61**

- C60a What are those signs and symptoms? ប្រសិនបើដឹង, តើរោគសញ្ញាទាំងនោះមានអ្វីខ្លះ? **(ចំណើយអាចមានច្រើន, កុំអានចំណើយ)**
- a [] Difficult/fast breathing ពិបាកដកដង្ហើម
 - b [] Fever គ្រុនក្តៅ
 - c [] Cough ក្អក
 - d [] Muscle Ache ឈឺសាច់ដុំ
 - e [] Sore throat ឈឺករ
 - f [] Other (Specify) ផ្សេងទៀត _____

C61 Do you believe you can protect your poultry and yourself from AI? 1 Yes 0 No 9 Don't know
 តើអ្នកជឿថា អ្នកអាចការពារ មាន់ទា របស់អ្នក និង ខ្លួនអ្នកផ្ទាល់ មិនឱ្យឆ្លងជំងឺគ្រុនផ្តាសាយបក្សី បាន ដែរឬទេ?

C62 Do you think touching sick/dead poultry can cause fever? 1 Yes 0 No
 តើអ្នកគិតថា ការប៉ះពាល់ មាន់ទា ដែលឈឺ ឬ ងាប់ដោយសារជំងឺ អាចបង្កអោយកើតគ្រុនក្តៅ ដែរឬទេ?

C62a Have you experience any fever after touching sick or dead (died from illness) poultry? 1 Yes 0 No **If NO, Go to D1**
 តើអ្នកធ្លាប់ កើតគ្រុនក្តៅ ដែរឬទេ ក្រោយពីបានប៉ះពាល់ មាន់ទា ដែលឈឺ ឬ ងាប់ដោយសារជំងឺ ?

C62b Did you seek medical treatment for your symptoms? 1 Yes 0 No
 បើមាន តើ នៅពេលនោះ អ្នកបានស្វែងរកការព្យាបាលនៅទីណា ដែរឬទេ ?

I'd like to thank you for taking the time to answer my questions. Your involvement in this study is very important. Please accept this token of our appreciation as a thank you. (Hand out compensation kit)

ខ្ញុំបានបញ្ចប់សំណួរទាំងអស់ហើយ ដូចនេះ ខ្ញុំសូមអរគុណ ដែលអ្នកបានចំណាយពេលវេលា ឆ្លើយសំណួរទាំងអស់នេះ ។
 ការចូលរួមផ្តល់ចំណេះដឹងរបស់អ្នក ពិតជាមានសារៈសំខាន់ណាស់ ។ ខ្ញុំសូមជូនកញ្ចប់នេះ ទុកជាវត្ថុអនុស្សាវរីយ៍ ។

OBSERVATION Interviewer: Please take a few moments and fill in the following observational survey.

ការសង្កេត សូមបំពេញចំណើយខាងក្រោម ដោយធ្វើការសង្កេតនៅក្នុងបរិវេណផ្ទះ

01 See any DUCKS in the property 1 Yes 0 No **If NO, Go to Q# 05**
 ឃើញមាន ទា នៅក្នុងទីធ្លា

02 Ducks in pond 1 Yes 0 No 9 Don't have pond គ្មានត្រពាំងទឹក
 ឃើញមាន ទា នៅក្នុងត្រពាំងទឹក

03 Ducks are free ranging 1 Yes 0 No
 ឃើញមាន ទា ដើរដោយសេរី ក្នុងផ្ទះ

04 Ducks have contact pigs 1 Yes 0 No
 ឃើញមាន ទា ទៅនៅលាយឡំជាមួយជ្រូក

05 See any CHICKENS in the property 1 Yes 0 No **If NO, Go to Q# 08**
 ឃើញមាន មាន់ នៅក្នុងទីធ្លា

06 Chickens are free ranging 1 Yes 0 No
 ឃើញមាន មាន់ ដើរដោយសេរី ក្នុងផ្ទះ

07 Chickens have contact with pigs 1 Yes 0 No
 ឃើញមាន មាន់ ទៅនៅលាយឡំជាមួយជ្រូក

08 See poultry scrap (feathers...) on the property 1 Yes 0 No
 ឃើញ មានសំណល់ពីធ្វើមាន់ទា នៅក្នុងទីធ្លា (ស្លាប...)

09 See poultry faeces on the property 1 Yes 0 No
 ឃើញលាមកមាន់ទា រាយប៉ាយ នៅក្នុងទីធ្លា

010 See any protective material (used or not used) in the property, as listed: ឃើញមានសម្ភារៈពាក់ការពារ (ទោះបី ឬ មិនប្រើ) នៅក្នុងផ្ទះ ដូចជា:
 a Gloves b Rubber boots c Masks d Apron e None
 ស្រោមដៃ ស្បែកជើងជ័រ ឃ្មុប ឬ ក វែង ម៉ាស់ អាវការពារ មិនឃើញមានអ្វីទាំងអស់

011 See anyone wearing protective material while caring for or preparing poultry for food 1 Yes 0 No
 ឃើញអ្នកកំពុងសំអាតទ្រូងមាន់ទា ហើយមានពាក់ សម្ភារៈពាក់ការពារ

**H5N1 KAP survey among backyard poultry farmers
Child Questionnaire**

កំរងសំណួរ សំរាប់កុមារ អាយុ ១៥ ឆ្នាំ ឬ តិចជាង

Today Date: ____/____/____

Interviewer's name _____

Village ភូមិ _____

Province ខេត្ត _____

GPS | | | | | | | | | | | | | | | |

Province Village Family Interviewed member
ID Code | | | | | | | | | | | | | | | | | | | | | |
(TK = Takeo, SV = Svey Rieng; PR=Pursat; BM=Banteay Meanchey; Exp. KC010105)

Instructions to Interviewer: ការណែនាំសំរាប់អ្នកសួរ

កំរងសំណួរនេះប្រើសំរាប់សួរ កុមារអាយុ ១៥ ឆ្នាំ ឬ តិចជាង ។ សូមអានគ្រប់សំណួរទាំងអស់ និង ធ្វើតាមការណែនាំក្នុងការលោតរំលងសំណួរ ទៅកាន់ផ្នែកផ្សេងៗ នៃកំរងសំណួរ ។
សូមត្រូវចំណីដល់សំណួរដែលបានសួរ និង សូមកុំរំលងសំណួរ បើមិនមានកំណត់សំគាល់ជាអោយរំលង ។
សូមសរសេរលេខកូដ នៅផ្នែកខាងលើ ខាងស្តាំ ទំព័រនីមួយៗរបស់កំរងសំណួរ ។ ការសរសេរលេខកូដនេះមានសារៈសំខាន់ណាស់ ព្រោះថាទំព័រមុខរបស់កំរងសំណួរ នឹងត្រូវផ្តាច់ចេញ ហើយទុកដោយឡែក មុននឹងធ្វើការបញ្ចូលទិន្នន័យ ។
ប្រសិនបើអ្នកមានចំងល់ផ្សេងៗទាក់ទងនឹងកំរងសំណួរ សូមទូរស័ព្ទទាក់ទងទៅ ឈ្មោះ ម៉ារ៉ូ ដែលមានលេខ 012 802 475 ឬ ទៅ ឈ្មោះ សុវត្ថិ ដែលមានលេខ 012 322 031 ។
ពេលដែលបំពេញកំរងសំណួរចប់សព្វគ្រប់ សូមទុករក្សាទុកកំរងសំណួរជាមួយគ្នាដោយចាត់ទៅតាមផ្ទះនីមួយៗ រហូតដល់ចប់ថ្ងៃនីមួយៗ ។ បន្ទាប់ពីនោះទើប ទំព័រមុខនឹងត្រូវផ្តាច់ចេញពីកំរងសំណួរ ។

F. Questions for children only សំណួរ សំរាប់កុមារ

Poultry Handling Behavior in Children ឥរិយាបថរបស់កុមារ ក្នុងការប៉ះពាល់មាន់ទា

Who is responding to this questionnaire? តើ នរណាជាអ្នកឆ្លើយនិង សំណួរទាំងនេះ? a child កុមារ b parent ឪពុក ឬ ម្តាយ

N.B. IF THE PARENT IS ANSWERING QUESTIONS: ចំណាំ បើសិនជា ឪពុកម្តាយឆ្លើយនិងសំណួរទាំងនេះ
ALL QUESTIONS REFER TO THE CHILD (e.g., Do you go to school? Means does the child go to school?)
រាល់សំណួរត្រូវសួរអំពីកុមារ (ឧទាហរណ៍ សំណួរសួរថា តើអ្នកចូលរៀនដែរទេ? មានន័យថា តើកុមារចូលរៀនដែរឬទេ?)

Read to respondent: I would like to ask you a few questions about your daily life. If you do not understand a question or I am speaking too fast, please let me know and I will repeat the question. Ok, lets begin:

ខ្ញុំចង់សុំសួរអ្នក នូវសំណួរខ្លះៗអំពីជីវិតប្រចាំថ្ងៃរបស់អ្នក។ បើសិនអ្នកមិនយល់ ឬ ខ្ញុំនិយាយលឿនពេក សូមអ្នកប្រាប់ខ្ញុំ ខ្ញុំនឹងសួរសំណួរនោះ ម្តងទៀត។
ខ្ញុំសុំសួរ ថា:

Child's Surname (only)
ឈ្មោះកុមារ _____

1 Sex of child ភេទរបស់កុមារ 1 Male ប្រុស 2 Female ស្រី

2 Age of child អាយុរបស់កុមារ _____ years old ឆ្នាំ

3 Do you go to school? តើអ្នកចូលរៀនហើយឬទេ? 1 Yes 0 No

4	Do you have poultry at home?	តើផ្ទះប្អូន មានចិញ្ចឹម មាន ទា ដែរឬទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	If NO, Go to Q#6	
5	Do you help take care of poultry?	តើអ្នកមានជួយថែទាំមាន ទា ដែរឬទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
Do you:						READ Q5a-5e
5a	Gather and place poultry in cages?	តើអ្នក មានជួយចាប់មាន់ទា ដាក់ក្នុងទ្រុងដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
5b	Feed poultry?	តើមានជួយអោយចំណីមាន់ទាដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
5c	Treat sick poultry with medicine?	តើអ្នកមានជួយរកថ្នាំមកព្យាបាលមាន់ទាលើដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
5d	Clean poultry faeces?	តើអ្នកមានជួយសំអាតទ្រុងមាន់ទាដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
5e	Touch eggs?	តើអ្នកមានជួយរើស ពងមាន់ ពងទា ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
6	Do you have fighting cocks at home?	តើផ្ទះប្អូន មានចិញ្ចឹម មាន់ជល់ ដែរឬទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
7	Do you care for fighting cocks?	តើអ្នកមានជួយមើលថែរ មាន់ជល់ ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	READ ALL Q7a-7g	
7a	Feed fighting cocks?	តើប្អូន មានជួយឱ្យចំណី មាន់ជល់ ដែរឬទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
7b	Touch bloody cocks?	តើអ្នកធ្លាប់បះពាល់ឈាមមាន់ជល់ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
7c	Blow the beak?	តើអ្នកធ្លាប់បិតជំពុះមាន់ជល់ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
7d	Kiss, Suck or lick wounds?	តើអ្នកធ្លាប់ បិត របូសមាន់ជល់ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
7e	Share water from the same bottle used for drinking or spraying?	តើអ្នកធ្លាប់បៀមទឹក ព្រួសអោយទឹកមាន់ជល់ ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
7f	Clean trachea with swab or feathers?	តើអ្នកធ្លាប់ត្បាញសំអាតបំពង់ក មាន់ជល់ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
7g	Clean feathers?	តើអ្នកធ្លាប់ជូត លាង សំអាតរោមមាន់ជល់ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
8	Do you wear protective equipments (gloves, boots, aprons) when you care for poultry?	តើអ្នកពាក់អ្វីការពារទេនៅពេលដែលអ្នកថែទាំមាន់ទា?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> If no poultry	
Do you wear:		តើអ្នកពាក់			If NO, Go to Q#9	
8a	gloves	ស្រោមដៃ	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	(សូមអានចំណើយ)	
8b	boots	ស្បែកជើងកវែង	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
8c	apron	ខោអាវការពារប្រឡាក់	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
8d	face mask	ពាក់ម៉ាស់មុខ	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
9	Do you collect dead birds from outside of your home?	ពេលប្រទះឃើញបក្សីងាប់នៅក្រៅផ្ទះ តើអ្នកដែលរើសយកមកផ្ទះដែរឬទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	If NO, Go to Q#10	
9a	Are these	តើជាបក្សីស្រុក ឬ បក្សីព្រៃ?	1 <input type="checkbox"/> domestic poultry	2 <input type="checkbox"/> wild birds	9 <input type="checkbox"/> Don't Know	
10	Do you remove feathers from birds?	តើអ្នកធ្លាប់បោចរោម មាន់ទា ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	If NO, Go to Q#11	
10a	Have you ever removed feathers from sick/dead birds?	តើអ្នកធ្លាប់បោចរោម មាន់ទា ដែលឈឺ ងាប់ដោយជំងឺ ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
11	Do you help prepare poultry for family meals?	តើអ្នកជួយធ្វើមាន់ទាសំរាប់ធ្វើម្ហូបដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	READ ALL 11a- 11g	
11a	Do you: Boil birds?	តើអ្នកមានជួយស្ទោរមាន់ទាដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
11b	Slaughter/bleed poultry?	តើអ្នកមានជួយអារក សំលាប់ មាន់ ទា ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
11c	Cut poultry meat?	តើអ្នកមានជួយ កាប់សាច់ មាន់ ទា ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
11d	Wash poultry meat?	តើអ្នកមានជួយ លាងសាច់ មាន់ ទា ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
11e	Remove internal organs from birds?	តើអ្នកមានជួយ រះយកគ្រឿងក្នុងមាន់ទាដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		
11f	Wash internal organs?	តើអ្នកមានជួយ លាងសំអាតគ្រឿងក្នុងមាន់ទាដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No		

12	Do you hunt/catch wild birds? តើអ្នកចាប់សត្វបក្សីព្រៃដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	If NO, Go to Q#13
12a	Do you catch wild birds with bare hands? តើអ្នកចាប់សត្វបក្សីព្រៃនិងដៃទេដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	
13	Do you:			
13a	Touch/Play with birds that are alive? តើអ្នកចាប់មាន់ទា (រស់) លេងដែរឬទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	
13b	Touch/Play with sick birds? តើអ្នកលេងជាមួយមាន់ទាឈឺដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	
13c	Touch/Play with dead birds? តើអ្នកមានរើស មាន់ទាងាប់ យកមកលេង ដែរឬទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	
13d	Attend cock fighting matches? តើអ្នកមានទៅមើលជល់មាន់ដែរទេ?	1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	
14	Do you have bath or swim in ponds where poultry have access to? តើអ្នកងូតរឺហែលទឹកក្នុងស្រះដែលមានមាន់ទាហែលលេងដែរទេ?			
		1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	If NO, Go to Q#15
14a	How many times per week? បើធ្លាប់ តើអ្នកងូតរឺហែលប៉ុន្មានដងក្នុង១អាទិត្យ?			
	1 <input type="checkbox"/> 1-3 ដង	2 <input type="checkbox"/> 3-5 ដង	3 <input type="checkbox"/> 5-6 ដង	4 <input type="checkbox"/> everyday រាល់ថ្ងៃ
15	Do you wash your hands after touching poultry? តើអ្នកលាងដៃបានរាល់ដង ដែរទេ ក្រោយពេលប៉ះពាល់មាន់ទារួច?			
		9 <input type="checkbox"/> If no poultry	1 <input type="checkbox"/> Always	2 <input type="checkbox"/> Sometimes
			0 <input type="checkbox"/> Never	If Never, Go to Q#16
15a	Do you use soap? តើអ្នកលាងជាមួយសាប៊ូ បានរាល់ដងឬទេ?			
		1 <input type="checkbox"/> Always	2 <input type="checkbox"/> Sometimes	0 <input type="checkbox"/> Never
Skip if parent is answering for the child បើអាណាព្យាបាលជាអ្នកឆ្លើយជំនួសកុមារ (ដែលក្មេងពេក) សូមបញ្ចប់កំរងសំណួរត្រឹមនេះ				
Attitudes				
16	Can you tell if poultry is sick/ill? តើអ្នកដឹងថា មាន់ទា ណាឈឺ មាន់ទា ណាមិនឈឺទេ?			
		1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> Don't Know If NO or DK, Go to Q#17
16a	If Yes, how can you see that a chicken or ducks is sick? មាន់ទា ពេលវាឈឺ តើវាធ្វើទុក្ខយ៉ាងម៉េចខ្លះ?			
			
			
			
17	Have you ever heard of bird flu? តើអ្នកធ្លាប់ឮអំពី ជំងឺផ្តាសាយបក្សី ដែរទេ?			
		1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	If NO, Go to Q#19
18	Where did you hear about bird flu? (check all that apply) តើអ្នកបានឮមកពីណាខ្លះ?			
	a <input type="checkbox"/> family សមាជិកគ្រួសារ	e <input type="checkbox"/> TV ទូរទស្សន៍		
	b <input type="checkbox"/> school/teacher សាលារៀនរឺគ្រូ	f <input type="checkbox"/> Village chief មេភូមិ		
	c <input type="checkbox"/> poster ផ្ទាំងរូបភាពអប់រំ	g <input type="checkbox"/> Village Vet បុគ្គលិកពេទ្យសត្វភូមិ		
	d <input type="checkbox"/> radio វិទ្យុ	h <input type="checkbox"/> others ផ្សេងៗ	_____	
19	Can you get bird flu? តើប្អូនគិតថា ខ្លួនប្អូន អាចឆ្លងជំងឺផ្តាសាយបក្សីពី មាន់ទា ដែរឬទេ?			
		1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	9 <input type="checkbox"/> Don't Know
20	How can you get bird flu? (open question) បើអាចឆ្លង តើប្អូនគិតថាវាឆ្លងមកប្អូន តាមណា, ឆ្លងយ៉ាងដូចម្តេច?			
			
			
			
21	How can you protect yourself from getting bird flu? តើប្អូន គួរធ្វើយ៉ាងដូចម្តេចខ្លះ ដើម្បីការពារកុំអោយឆ្លងជំងឺផ្តាសាយបក្សី មកខ្លួនប្អូនបាន?			
			
			
			

Appendix C: Interviewer Training Manual



INSTITUT PASTEUR
DU CAMBODGE

Phase 1: November – December 2006
Phase 2: November – December 2007

Interviewer Training Manual

KNOWLEDGE, ATTITUDES AND PRACTICES (KAP) SURVEY OF BACKYARD POULTRY OWNERS AND THEIR FAMILIES

Instructions

You have been hired to participate in a KAP survey of backyard poultry owners and their families. Your principal role is to conduct interviews with the study participants. Your role is very important for this study.

The purpose of the survey is to evaluate poultry handling behaviors of backyard poultry owners and their understanding of avian influenza (“bird flu”). Your task is to interview the subjects included in this study. The quality of your data collection is directly responsible for the quality of the data that will be used in this study.

You will be working in teams and your team responsibilities include:

- Inform village chief of purpose of study and obtain permission to include his village
- Identify households in village (5-6 households per village) to include in study
- Data collection: interview subjects
- Keep study files in order
- Maintain appropriate time schedule
- Report questions/problems to supervisor
- Provide feedback to supervisor

This manual provides details of your responsibilities. Please read this manual carefully. You can refer to this throughout the duration of the study if you have questions. You may also contact the supervisors at:

Maria Van Kerkhove
012 802 475

Sowath Ly
012 322 031

Please enter your information here:

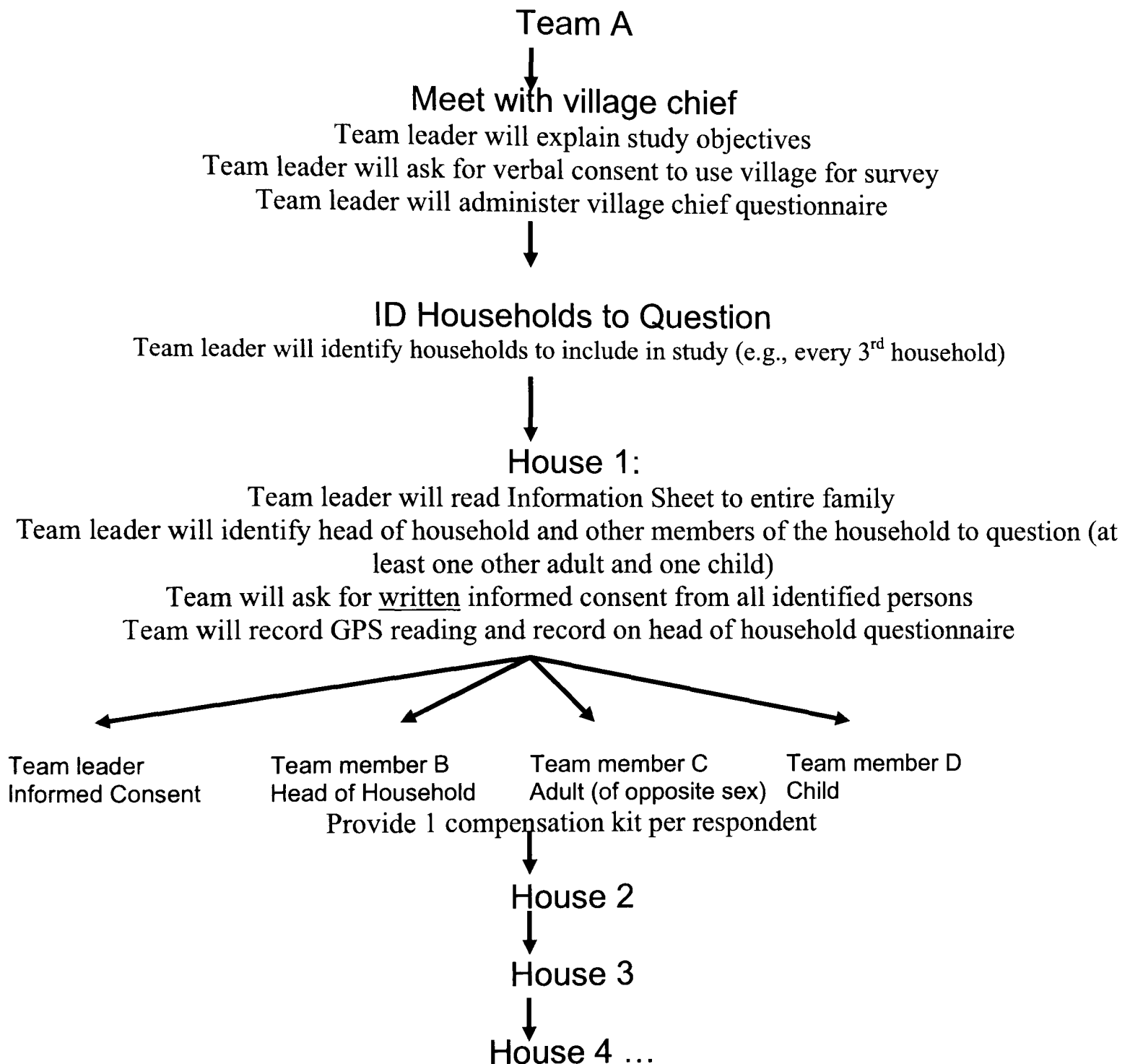
Name: _____

Phone number: _____

Email address: _____

Procedures

You will be working in teams of 5 (1 team leader, 3 interviewers and 1 local guide). Each team will be provided a car and driver. Your role as a team is to administer the questionnaires to thirty people per village per day. Below is an outline of your procedures for each village your team visits. You and your team will be responsible for completing 5 villages per week (1 village [30 interviews] per day). Each step is described in detail below this flowchart.



At the end of the day, team leader will pay compensation to local guide (\$5)
At the end of each day, you must have completed 10 male and 10 female adults (5 head of household and 15 adult) and 10 children

Procedures in detail:

Step 1: Meet with village chief, explain study objectives to the village chief, get verbal consent to include his village, administer the “village chief” questionnaire

Each morning, you and your team will be driven to your assigned village. You must first find and meet with the village chief or vice village chief. Prior to your visit, the village chief will be contacted by IPC staff who will provide background of the study and arrange your visit.

The team leader will explain to the village chief that we would like to include their village in this KAP study of poultry handling behavior. In your explanation, you should include the following information:

- We would like to include 30 people from his village in the study,
- We will need one day to complete the questionnaires,
- There are no risks to his villagers to participate in the survey, and
- That we will compensate each participant for his or her time. Compensation includes a kit of thick rubber gloves and a bar of soap (worth approximately \$1).

Once the village chief agrees, the team leader will administer the VILLAGE CHIEF QUESTIONNAIRE (Appendix A). This is a short, one-page questionnaire. Ask the village chief to sketch an outline of the village, identifying roads, location of houses and boundaries of village.

Ask the village chief if he would like to accompany you as the local guide for the day. If he cannot, ask if he can provide a local guide. The local guide will be compensated \$5/day.

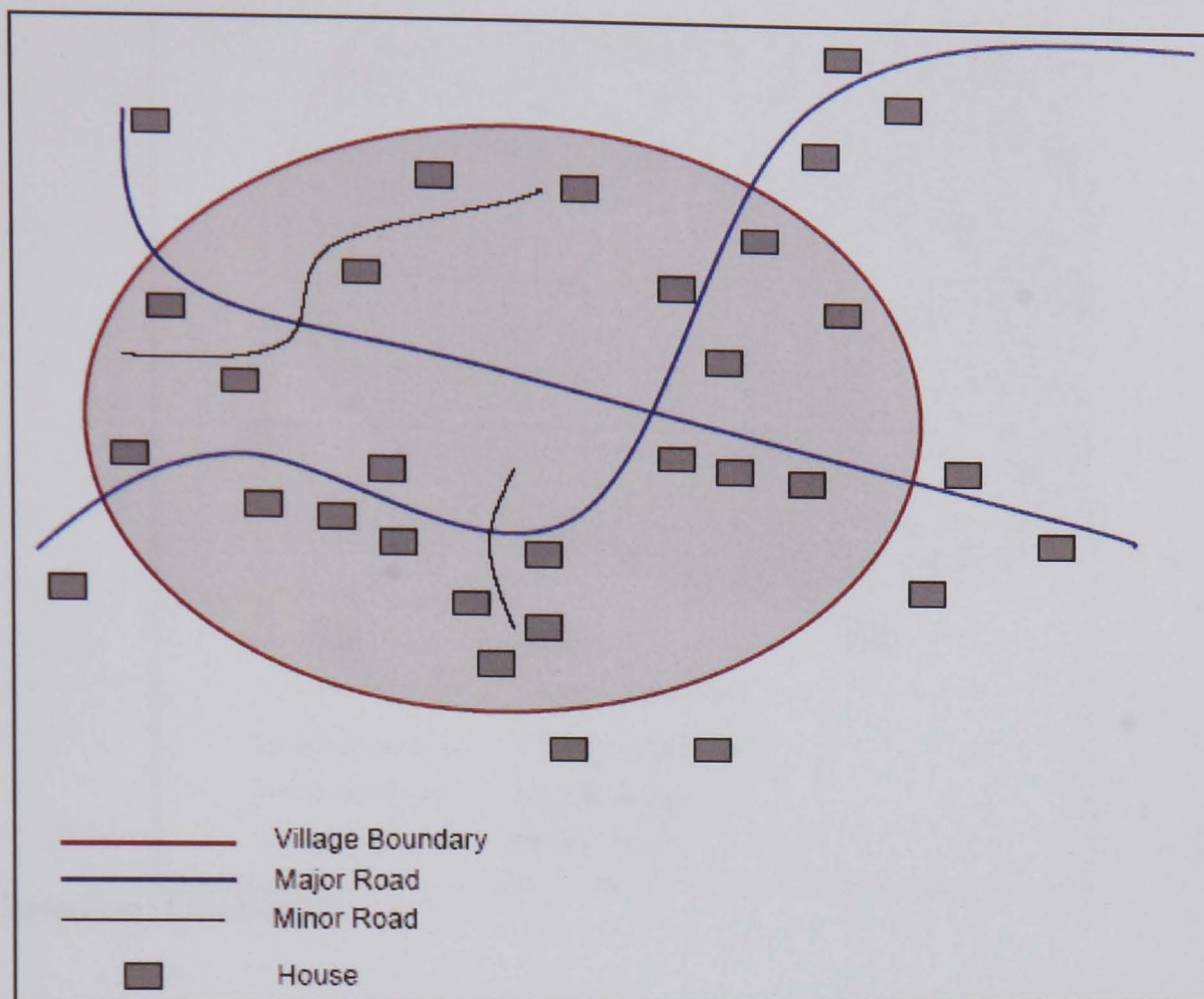


Figure 1. Example sketch of village

Step 2: Identify households to include in study

After you've obtained verbal consent from the village chief and administered the VILLAGE CHIEF QUESTIONNAIRE, ask your local guide to bring you to the center of the village. From the center of the village, choose a direction to work through the village by spinning a pen on a flat surface. In that direction, you will choose every N^{th} house.

The sampling plan for:

- Team A – include every 9th house
- Team B – include every 3rd house
- Team C – include every 6th house
- Team D – include every 9th house

Using the example of the village sketch shown in Figure 1, we can show movement of Team B in orange. Houses are numbered from the center of the village, House 1, House 2, House 3, etc. Households 3, 6, 9, 12, etc (colored in red) should be included in the study.

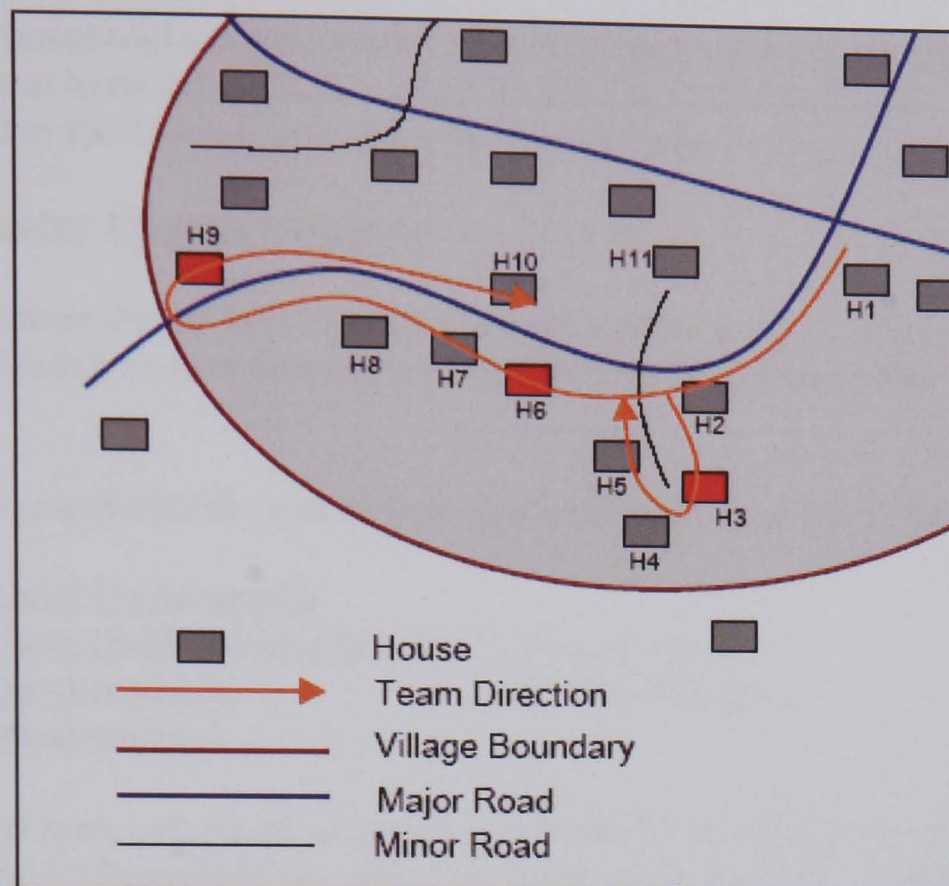


Figure 2. Selection of households

Within each village you must complete a questionnaire for 10 adult (16 yrs old and older) males, 10 adult (16 yrs old and older) females and 10 children (15 and younger).

- For example, if you interview 6 people/household, you must visit 5 households to complete 30 interviews.
- For example, if you interview 5 people/household, you must visit 6 households to complete 30 interviews.

Step 3: Obtain informed consent from household participants

Go as a team to your first household.

Team Leader:

Parents or guardians (e.g., grandparents) must be at home. If parents or guardians are not home, go to the next assigned household. The team leader will read the information sheet (Appendix B) to the entire family and identify the head of household and *at least four* other household members (adults and children) to include in the study. After you read the information sheet to the family, have each participant sign the informed consent form. If the participant is 17 years old or younger, the parent or guardian must sign the consent form for the participant. **Each study participant must have a signed informed consent form before the interview takes place.**

Once signed, store all informed consent forms in the team leaders binder.

If members of the household are not home at the time of the initial visit, the team leader should make an appointment to revisit the house when the other residents are home. These people should be included in the 30 total interviews that you complete in your village.

Step 4: Administer Questionnaires

At each house, the team should interview all residents that are present. These should include: 1 head of household (adult male or female), and any adults (either gender) and children (boy or girl).

The team leader is responsible for making sure that at the end of each day, your team has completed:

- (1) 1 Village Chief Questionnaire
 - (2) 5 Head of Household Questionnaires
 - (3) 15 Adult Questionnaires
 - (4) 10 Child Questionnaires
- } 10 Adult Male
} 10 Adult Female

Team leader: *Your team only needs to complete 5 Head of Household Questionnaires per day. If you visit more than 5 households per day, you only need to interview adults and children at the remaining households to reach your goal of 31 interviews per village.*

Four questionnaires have been created for this survey. Below is a description of each questionnaire:

1- Village Chief Questionnaire (Appendix C)

A one-page questionnaire should be filled out by the team leader. These short questionnaires aim to collect basic information about the village (e.g., number of households, nearest health center) and data on middlemen that visit the village. The team leader will fill out 1 village chief questionnaire per village.

2- Head of household questionnaires (Appendix D)

This questionnaire is intended for the head of the household (an adult male or female) living in the house. This questionnaire is long and is intended for only ONE (1) family member per household. This questionnaire contains questions on addressing questions of poultry ownership, raising, mixing, movement (trading, selling and transport), fighting cock ownership, extent of wild bird mixing; poultry handling behavior, mortality reporting, knowledge and attitudes about AI. One interviewer (Interviewer #1) will be responsible for administering the head of household questionnaire only (5/day).

3- Adult family member, not head of household, who is 15 years old or older (Appendix E)

This questionnaire is intended for all other adults (not the head of household) residing in the household. This questionnaire contains some of the same questions as the “head of household questionnaire” however it is shorter in length. The questionnaire contains questions addressing poultry handling behavior, mortality reporting, knowledge and attitudes about AI. The

GPS Coding

With your GPS meter, record the coordinates at the front door of the house you are visiting. These coordinates should be written on the questionnaire in the box indicated below. Be sure to record these coordinates on the head of household questionnaire.

GPS

Procedures for asking questions in structured interviews

Your role as an interviewer is to conduct structured interviews with backyard poultry owners. You need to ensure uniform answers from ALL respondents. We want to make sure that all interviews are conducted exactly the same way. Steps 1-8 below provide instructions on how to conduct structured interviews.¹

- 1) Read the questions exactly as they are worded in the questionnaire
- 2) Read each question slowly
- 3) Ask the questions in the order that they are presented in the questionnaire
- 4) Ask every question that applies to the respondent (all questions that are not appropriate for the respondent will be indicated with “skip” instructions)
- 5) Repeat the whole question if it is misheard or misunderstood
- 6) Use only allowable probes. For example:
 - a. If a respondent does not answer a question completely, you can ask “What do you mean exactly?” or say “I don’t think that I understand.” “Could you explain that a little?”
 - b. Other allowable probes include: For questions that ask to define quantity, if the respondent is too vague (e.g., once or twice a month) you can ask “Can you be more exact?” For open-ended questions, if you feel the respondent has provided an incomplete answer, you can ask “Is there anything else that you can think of?”
- 7) Do not add apologies or explanations for questions unless they are printed in the questionnaire
- 8) Provide feedback to the respondent. For example, say “thank you,” “uh-huh” or “ok” while the respondent is answering questions.

¹ Adapted from Armstrong BK, White E and Saracci R. Principles of Exposure Measurement in Epidemiology, Monographs on Epidemiology and Biostatistics, Oxford University Press, 1995.

Step 5: Review and store completed forms

Before you and your team leave the household, all team members should take a few minutes to look over their completed questionnaires. The team leader is responsible to ensure that:

- There is an answer for EVERY question that is appropriate for the respondent in the questionnaire. If you do not, you will need to re-ask the participant.
- The participants ID code is written in the top right hand corner of each page of their questionnaire.
- The GPS code for the household is written on the front page of each questionnaire filled out at that household.

All completed forms (consent forms, questionnaires) should be stored in the “COMPLETED FORMS” binder. Keep all forms for each household together and clip together with a paper clip.

Maintaining interviewing timing

The team leader is responsible for maintaining time throughout the day. It will be essential to keep your team on schedule throughout the data collection. Your team will be responsible for completing 5 villages per week; this means completing 1 village (30 interviews) each day. We only have one week to complete each study area.

Check List:

Below is a list of the tasks you must complete during each household visit.

Check when complete	Description of task
Obtain informed Consent	
	The village chief has given verbal consent
	The team leader has read the information sheet to all household members and answered any questions of the family
	I have a signed informed consent sheet from ALL participants in the household
	I have filed all signed informed consent sheets in the team leaders binder
	Make appointment for household members not at home during initial visit
Fill out questionnaire	
	I have written my initials in the space provided at the top of the questionnaire
	I have assigned an ID number to each participant
	I have written the ID number on the top of each page of the participants questionnaire
	I have recorded the GPS coordinates for the house and written the coordinates on each participant from this household
	Interviewer 1 has administered a head of household questionnaire
	Interviewer 2 has administered a household adult questionnaire #1
	Interviewer 3 has administered a child questionnaire #1
	Interviewer 2 has administered a child questionnaire #2
	Interviewer 3 has administered a household adult questionnaire #2
	The team has completed 5 questionnaires from this house
	I have given a compensation kit to each participant
Checking the questionnaire	
	All questionnaires have been checked for completeness
Storing questionnaires	
	I have stored all completed questionnaires in my binder
At the end of each day:	
	My team has completed 31 interviews , including <ul style="list-style-type: none"> • 10 adult males > 15 years old • 10 adult females >15 years old • 10 children ≤ 15 years old • 1 village chief • Team leader, provide compensation (\$5) for local guide

KNOWLEDGE, ATTITUDES AND PRACTICES (KAP) SURVEY OF BACKYARD POULTRY OWNERS AND THEIR FAMILIES

Contract For Interviewer

I _____ agree to work as an interviewer in the Knowledge, Attitudes and Practices (KAP) Survey of Backyard poultry owners and their families. I understand my responsibilities described in the training manual. By signing this contract, I agree to:

1. Work as a team to interview 30 adults and children in one village each day
2. To complete 5 villages per week
3. To work all four weeks of the survey
 - a. Kampong Cham: 28 November – 3 December 2007
 - b. Rest day: 4 December
 - c. Prey Veng: 5 December – 9 December 2007
4. I understand that I will be paid \$15/day; including travel days and that I will be paid half of the amount earned each week of the survey (\$45). My remaining salary will be paid at the *completion* of data collection.
5. The cost of travel will be taken care of by IPC.
6. I understand that the cost of food and guest house is my own responsibility

Signed

Name Printed:
Interviewer
Date: 21 Nov 2007

Maria Van Kerkhove
Institut Pasteur du Cambodge
Date: 21 Nov 2007

Appendix D: Consent Forms in English and Khmer

Appendix D

ការសិក្សាពី ចំណេះដឹង, ឥរិយាបថ និង ការអនុវត្ត (KAP)

ព័ត៌មានសំរាប់អ្នកចូលរួមក្នុងការសិក្សា

យើងខ្ញុំសូមអោយអ្នកចូលរួមក្នុងការសិក្សាស្រាវជ្រាវអំពីជំងឺគ្រុនផ្តាសាយបក្សី ។ គោលបំណងនៃលិខិតយល់ព្រមនេះគឺ ផ្តល់ជូនលោកអ្នក នូវព័ត៌មានដែលលោកអ្នកត្រូវការដើម្បីជាជំនួយក្នុងការសំរេចចិត្ត ថាតើគួរចូលរួមក្នុងការសិក្សានេះឬអត់ ។ យើងខ្ញុំនឹងអានលិខិតយល់ព្រមនេះអោយអ្នកស្តាប់ ។ អ្នកក៏គួរតែអានលិខិតយល់ព្រម និង ព័ត៌មាននេះដោយយកចិត្តទុកដាក់ ។ លោកអ្នកអាចសួរយើងខ្ញុំបានទាក់ទងនឹងគោលបំណង នៃការសិក្សាស្រាវជ្រាវ អ្វីដែលយើងខ្ញុំចង់អោយអ្នកធ្វើ គ្រោះថ្នាក់ និង ផលប្រយោជន៍ដែលលោកអ្នកទទួលបានសិទ្ធិរបស់លោកអ្នកក្នុងនាមជាអ្នកស្ម័គ្រចិត្ត និងអ្វីផ្សេងទៀតទាក់ទងនឹងការស្រាវជ្រាវ ឬ លិខិតនេះដែលលោកអ្នកមិនទាន់យល់ច្បាស់ ។ នៅពេលគ្រប់សំណួររបស់អ្នកត្រូវបានឆ្លើយតបហើយ លោកអ្នកអាចសំរេចចិត្តបានថាគួរចូលរួមក្នុងការសិក្សានេះ ឬ អត់ ។ ម្យ៉ាងវិញទៀត យើងខ្ញុំចង់អោយអ្នកដឹងថា យើងខ្ញុំបានទាក់ទងទៅប្រធានភូមិរបស់អ្នកហើយ ហើយគាត់បានយល់ព្រមអោយយើងខ្ញុំធ្វើការសិក្សានៅក្នុងភូមិរបស់អ្នក ។

អាស័យដ្ឋានទាក់ទងរបស់អ្នកអង្កេត:

ម៉ារីយ៉ា វ៉ាន់ ឃើកយូរ

វិទ្យាស្ថានប៉ាស្ទ័រកម្ពុជា,

ផ្នែក អេពីដេមីយ៉ូសាស្ត្រ

ភ្នំពេញ, កម្ពុជា

maria@pasteur-kh.org

ទូរស័ព្ទ: (៨៥៥) ១២ ៨០២ ៤៧៥

ហេតុផលនៃការសិក្សា:

យើងខ្ញុំសូមអោយអ្នកចូលរួមក្នុងការសិក្សាដើម្បីរកអោយឃើញពីរបៀបដែលមនុស្សពេញវ័យ និង កុមារ មានទំនាក់ទំនង ជាមួយបសុបក្សី (មាន, ទា...) នៅតាមផ្ទះ ។ ប្រសិនបើ អ្នកចង់ចូលរួម គេនឹងសួរអ្នកទាក់ទងនឹងការចិញ្ចឹមបសុបក្សី, ឥរិយាបថ នៃការគ្រប់គ្រងបសុបក្សី, ការរាយការណ៍ពីការស្លាប់របស់បសុបក្សី និង ចំនេះដឹង និងឥរិយាបថ ចំពោះមុខ ជំងឺគ្រុនផ្តាសាយបក្សី ។

អ្នកមិនមានគ្រោះថ្នាក់ទាំងអស់ក្នុងការចូលរួមក្នុងការសិក្សានេះ ។

ប្រសិនបើអ្នកមានសំនួរអំពីការសិក្សានេះ អ្នកអាចសួរក្រុមអ្នកធ្វើការសិក្សានៅពេលដែលពួកគេចុះជួបអ្នក ។

សិទ្ធិក្នុងការបដិសេធ ឬ ដកខ្លួនចេញ:

អ្នកមានសិទ្ធិក្នុងការចូលរួម ឬ មិនចូលរួមក្នុងការសិក្សានេះ ។ បើសិនជាអ្នកជ្រើសរើសចូលរួមក្នុងការសិក្សា នេះ អ្នកមានសិទ្ធិដកខ្លួនចេញនៅពេលក្រោយ ។

ការរក្សាការសំងាត់

ព័ត៌មានផ្ទាល់ខ្លួនរបស់អ្នក នឹងរក្សាទុកជាសំងាត់ និង មិនចែកចាយដល់ អ្នកណាម្នាក់ក្រៅពីអ្នកអង្កេតចំបង ។ វិទ្យាស្ថានប៉ាស្ទ័រកម្ពុជានឹងបង់លេខនៅលើកំរងសំនួរដែលអ្នកបានឆ្លើយ និង រក្សាព័ត៌មាននេះនៅកន្លែងដែល មានសុវត្ថិភាព ។ រាល់ព័ត៌មានដែលយើងខ្ញុំប្រមូលបានពីអ្នក នឹងត្រូវបង់ លេខដូចលេខនៅលើកំរងសំនួរ ។ យើងខ្ញុំ នឹងមិនអាចដឹងថាលេខណាមួយជាលេខរបស់អ្នកទេ ។

តំលៃ/ការចំណាយក្នុងការចូលរួម

មិនមានការបង់ប្រាក់ទេក្នុងការចូលរួមក្នុងការសិក្សានេះទេ ហើយអ្នកនឹងមិនទទួលបាននូវកំរៃអ្វីពីការចូលរួម ក្នុងការសិក្សានេះទេ គឺមានតែ “កញ្ចប់ឧបត្ថម្ភ” រួមមាន ស្រោមដៃជ័រ និង សាប៊ូដុំ ដែលនឹងប្រគល់អោយអ្នកជាការតបស្នងចំពោះការចំណាយពេលវេលារបស់អ្នក ។

ក្រុមសីលធម៌

គណៈកម្មាធិការក្រុមសីលធម៌កម្ពុជា ក្រសួងសុខាភិបាល និង គណៈកម្មាធិការក្រុមសីលធម៌ របស់ មហា វិទ្យាល័យ London School of Hygiene and Tropical Medicine បានយល់ព្រមអនុញ្ញាតិ ឱ្យធ្វើការសិក្សានេះហើយ ។

**ការសិក្សាពី ចំណេះដឹង, ឥរិយាបថ និង ការអនុវត្ត (KAP)
លទ្ធផលប្រមូល**

ខ្ញុំបានអានព័ត៌មានដែលទាក់ទងនឹងការសិក្សានេះ ឬ បានស្តាប់ការពន្យល់ និង យល់ពីអ្វីដែលគេត្រូវការពីខ្ញុំ និង ពីអ្វីដែលនឹងកើតមានដល់ខ្ញុំ នៅពេលដែលខ្ញុំចូលរួមក្នុងការសិក្សានេះ ។

គ្រប់សំណួររបស់ខ្ញុំត្រូវបានឆ្លើយដោយបុគ្គលិករបស់គំរោង ។

ខ្ញុំដឹងថា ខ្ញុំអាចដកខ្លួនពីការសិក្សានេះពេលណាក៏បានដោយពុំចាំបាច់មានបញ្ជាក់ពីហេតុផល ។

អ្នកចូលរួមអាយុ ១៨ ឆ្នាំ ឬ លើស:

ខ្ញុំយល់ព្រមចូលរួមក្នុងការសិក្សានេះ ។ ខ្ញុំយល់ថាការចូលរួមក្នុងការសិក្សានេះជាជំរើសរបស់ខ្ញុំ ។ ខ្ញុំដឹងថា ខ្ញុំអាចបដិសេធមិនចូលរួមក្នុងការសិក្សានេះពេលណាក៏បានដោយមិនមានទទួលការពិន័យ ។

.....
ហត្ថលេខារបស់អ្នកចូលរួម ថ្ងៃខែឆ្នាំ

សំរាប់កុមារអាយុក្រោមអាយុក្រោម ១៨ ឆ្នាំ:

ខ្ញុំយល់ព្រមអោយកូនខ្ញុំចូលរួមក្នុងការសិក្សានេះ ។ ខ្ញុំយល់ថាការចូលរួមក្នុងការសិក្សានេះជាជំរើសរបស់ខ្ញុំ ។ ខ្ញុំដឹងថា ខ្ញុំអាចបដិសេធមិនអោយកូនខ្ញុំចូលរួមក្នុងការសិក្សានេះពេលណាក៏បានដោយមិនមានទទួលការពិន័យ ។

.....
ហត្ថលេខារបស់ឪពុកម្តាយ ឬ អាណាព្យាបាល ថ្ងៃខែឆ្នាំ

.....
ឈ្មោះរបស់កូន (សរសេរ)

.....
ហត្ថលេខារបស់អ្នកអង្កេត ថ្ងៃខែឆ្នាំ

Knowledge Attitudes and Practice (KAP) Survey INFORMATION SHEET FOR PARTICIPANTS

Investigator's statement:

We are asking you to be in a research study about bird flu. The purpose of the consent form is to give you the information you need to help you decide whether or not you want to participate in the study. We will read the consent form to you. You should also read the Information Sheet and the Consent Form carefully. You may ask questions about the purpose of the study, what we are asking you to do, the possible risks and benefits from the study, your rights as volunteer, and anything else about the study or the Consent Form that is not clear. When all your questions have been answered, you can decide whether you want to participate. This process is called "informed consent". In addition, we also would like you to know that your village Chief has been contacted. He approved that we conduct the study in your village among those who are willing to participate.

Contact details of investigator:

Maria Van Kerkhove
Institut Pasteur du Cambodge
Epidemiology Unit
Phnom Penh, Cambodia
maria@pasteur-kh.org
Mobile: (855) 12 802 475

Reason for the Study:

You are being asked to participate in a study to find out how adults and children interact with backyard poultry. If you wish to participate, you will be asked a series of questions about poultry ownership and raising practices, poultry handling behavior, poultry mortality reporting, and knowledge and attitudes about bird flu.

There are no risks to joining this study.

If you have any questions throughout the study, you can ask the study team when they visit you.

Right to Refuse or Withdraw:

You are free to join the study or not. If you choose to join the study, you are free to drop out later. If you do not join the study, you can still get medical treatment as you have done in the past.

Confidentiality:

We will not record your name on the questionnaires. Personal information and information related to your behavior will be kept private and will not be shared with anyone other than the principal investigator. The Institut Pasteur, Cambodia will assign a number to your questionnaire and will keep this information in a secure place. Any facts that we collect about you will be labeled with this number. We will not have any way of knowing what number is assigned to you.

Cost / payment to participate:

There is no cost to you for participating in the study. You will not be paid for joining the study but a "compensation kit" containing rubber gloves and soap will be provided to you as a compensation of your time.

Ethical Considerations:

The Cambodian Ethical Committee, Ministry of Health and the London School of Hygiene and Tropical Medicine Ethical Committee have approved this study.

Knowledge Attitudes and Practice (KAP) Survey CONSENT FORM

I have read the information sheet concerning this study or have understood the verbal explanation and understand what will be required of me and what will happen to me if I take part in the study.

My questions have been answered by project staff.

In understand that at any time I may withdraw from this study without giving a reason.

PARTICIPANTS 18 YEARS OLD AND OLDER

I AGREE TO BE PART OF THIS STUDY. I UNDERSTAND THAT BEING PART OF THIS STUDY IS MY CHOICE. I UNDERSTAND THAT I CAN REFUSE TO BE PART OF THE STUDY AT ANY TIME WITHOUT PENALTY.

Parents or guardians' signature

Date

FOR CHILDREN UNDER 18 YEARS OF AGE:

I AGREE FOR MY CHILD TO BE PART OF THIS STUDY. I UNDERSTAND THAT BEING PART OF THIS STUDY IS MY CHOICE. I UNDERSTAND THAT I CAN REFUSE TO HAVE MY CHILD BE PART OF THE STUDY AT ANY TIME WITHOUT PENALTY.

Parents or guardians' signature

Date

Child's Name (printed)

Investigator signature

Date


**INSTITUT PASTEUR
DU CAMBODGE**



Unicef logo

Appendix E: Ethics Approval Forms from LSHTM and the Cambodian Ministry of Health

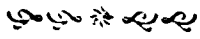


ក្រសួងសុខាភិបាល

MINISTRY OF HEALTH

គណៈកម្មាធិការជាតិក្រមសីលធម៌

អង្គការស្រាវជ្រាវសុខភាពដែលទាក់ទងនឹងបទុស្ស
National Ethics Committee for Health Research



លេខ... ១០៧... NECHR.

ព្រះរាជាណាចក្រកម្ពុជា
KINGDOM OF CAMBODIA
ជាតិ សាសនា ព្រះមហាក្សត្រ
NATION RELIGION KING



រាជធានីភ្នំពេញ, ថ្ងៃទី ០៧ ខែ ១១ ឆ្នាំ ២០០៦

Dr. Sirenda Vong
Principal Investigator

Project: Knowledge, Attitudes and practice survey to evaluate poultry handling behaviour among backyard poultry owners and their families and poultry market merchants : a cross-sectional survey of 4 geographic areas

Reference: November 3rd, 2006 NEC meeting minute

Dear Dr. Sirenda Vong,

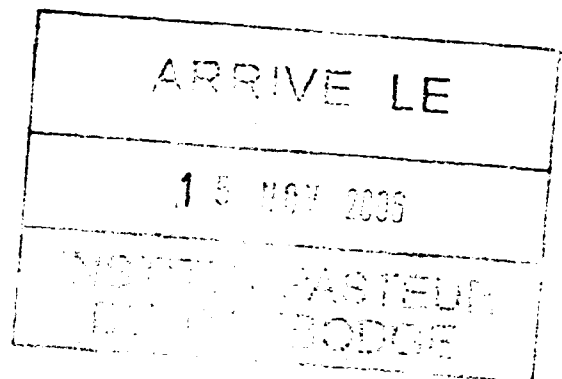
I am please to notify you that your project entitled "Knowledge, Attitudes and practice survey to evaluate poultry handling behaviour among backyard poultry owners and their families and poultry market merchants: a cross-sectional survey of 4 geographic areas" has been approved by the National Ethics Committee for Health Research in the meeting on November 3rd, 2006.

The principal investigator of the project shall also submit a copy of the progress and final report to the committee's secretariat at the National Institute of Public Health at #2 Kim Il Sung Blvd.. Khan Tuol Kok, Phnom Penh, Cambodia.(Tel: 855-23-880-345. Fax: 855-23-880-346)

Regards,

Chairman

H.E. Prof. ENG HUOT





ETHICS COMMITTEE

APPROVAL FORM

Application number: 5064

Name of Principal Investigator Maria Van Kerkhove

Department Epidemiology and Population Health

Head of Department Pat Doyle

Title: The epidemiology of highly pathogenic avian influenza in Cambodia:
Evaluating the movement of poultry and the extent of interaction
between poultry and humans as measures of the risks of sustained
transmissions in poultry and onward transmission to humans

Approval of this study is granted by the Committee.

Chair T. W. Meade
Professor Tom Meade

Date 20 November 2006

Approval is dependent on local ethical approval having been received.

Any subsequent changes to the consent form must be re-submitted to the
Committee.

Appendix F: Questionnaires for Poultry Traders

Market Sellers

Middlemen

H5N1 Survey of Wet/Live Market Merchants

Today Date: ____/____/____

Interviewer's name: | | | | |

Questions for wet market/ live market workers

Name of Market _____

Village _____

Commune _____

District _____

Province _____

Province Village Market Subject #
 ID Code | | | | | | | | | | | | | | | |
 (TK = Takeo, SV = Svey Rieng; PR=Pursat; BM=Banteay Meanchey)

GPS | | | | | | | | | | | | | | | |

Instructions to interviewer

The following series of questions are to be filled out by you through observations of the market. You must obtain informed verbal consent before observations are recorded.
 Do not record the respondent's name anywhere on the questionnaire.
 Write the ID Code at the top of each page of this questionnaire in the space provided (upper right hand corner of the page).

Observational Questions: Market Level		Alive	Dead	Whole	Part	Organs
1. What animals are sold at this market?	<input type="checkbox"/> Chickens	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Geese	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Singing birds	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Fighting cocks	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Ducks	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Pigs	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Cows	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Fish	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Other animals	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
1a. How many stalls are selling poultry at this market?	_____ stalls					
2. Are multiple species of birds caged together at this market?				1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	
3. How are birds stored at this market?	1 <input type="checkbox"/> Caged 2 <input type="checkbox"/> Tied together			3 <input type="checkbox"/> Poultry is already dead		
4. Are ANY birds roaming freely at this market?				1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	
5. Are there faeces on the ground at the market?				1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	

The remaining questions should be answered at the STALL level, not the market level.

Location of Stall

6. Identification features of Stall (location)

Observational Questions: Stall Level		Alive	Dead	Whole	Part	Organs
7. What animals are sold at this stall?	<input type="checkbox"/> Chickens	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Geese	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Singing birds	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Fighting cocks	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Ducks	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Pigs	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Cows	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Fish	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
	<input type="checkbox"/> Other animals	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
8. Are multiple species of birds kept together at this stall?				1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	
9. How are birds stored at this stall?	1 <input type="checkbox"/> All Caged 2 <input type="checkbox"/> Free roaming			3 <input type="checkbox"/> Poultry is already dead		
	4 <input type="checkbox"/> Some caged/some free roaming			5 <input type="checkbox"/> Tied together		
9a. If caged, are there trays to catch faecal matter underneath each cage?				1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	
10. Are there faeces on the ground at the stall?				1 <input type="checkbox"/> Yes	0 <input type="checkbox"/> No	

Instructions to Interviewer:
 The following questions are to be asked directly to the stall worker. Assure the merchant that the answers to the following questions will be kept confidential. Their name will not appear anywhere on this questionnaire and their information will not be shared with anyone without their consent.

11. Respondent: 1 Male 0 Female
 12. Age _____ years old
 13. How long have you been trading at this market?
 1 less than 1 yr 2 Between 1-2 yrs 3 Between 2-3 yrs 4 More than 3 yrs
 14. What type of trade do you do at this market?
 1 Sell poultry only 1 Buy poultry only 1 Sell & buy poultry

15. How many people are working at the stall today?
 15a How many people are responsible for preparing poultry for sale?
 Male Female

How many M/F are responsible for each task?

Is his done at the market or at home?
 15b Boiling home market
 15c Bleeding home market
 15d Defeathering home market
 15e Removing internal organs home market
 15f Butchering home market
 15g Evisceration before selling? home market

16. Are birds boiled (scalded) before defeathering? 1 Yes 0 No 9 DK
 17. Are there separate areas for each of the above tasks? 1 Yes 0 No 9 DK
 18. Are the merchants wearing
 18a gloves 1 Yes 0 No
 18b boots 1 Yes 0 No
 18c aprons 1 Yes 0 No
 18d face mask (plastic) 1 Yes 0 No
 18e face mask (cotton) 1 Yes 0 No
 18f other 1 Yes 0 No
 19. Are equipment cleaned after each use (after each bird)? 1 Yes 0 No 9 DK
 20. Are butchering surfaces cleaned after each use (after each bird)? 1 Yes 0 No 9 DK
 21. Is there a separate area for slaughtering and selling? 1 Yes 0 No 9 DK
 22. How are carcasses and other waste disposed of? _____ 9 DK

23. How many times a week do you clean your poultry cages/areas? _____ times a week 99 Never
 23a When you clean, do you use disinfectant? 1 Yes 0 No

24. How many middlemen do you buy from? _____ 0 if the obtain poultry themselves
 24a Where do they/you bring poultry from? 9 DK

	Province	District	Village
1 <input type="checkbox"/>	Vietnam	_____	_____
2 <input type="checkbox"/>	Thailand	_____	_____
3 <input type="checkbox"/>	Cambodia	_____	_____
4 <input type="checkbox"/>	Cambodia	_____	_____

25. Where were your chickens last purchased from?

	Province	District	Village
1 <input type="checkbox"/>	Vietnam	_____	_____
2 <input type="checkbox"/>	Thailand	_____	_____
3 <input type="checkbox"/>	Cambodia	_____	_____
4 <input type="checkbox"/>	Cambodia	_____	_____

26. Where were your ducks last purchased from?

	Province	District	Village
1 <input type="checkbox"/>	Vietnam	_____	_____
2 <input type="checkbox"/>	Thailand	_____	_____
3 <input type="checkbox"/>	Cambodia	_____	_____
4 <input type="checkbox"/>	Cambodia	_____	_____

H5N1 Survey of Middlemen

Page 4

Sketch poultry movement of subject

A large, empty rectangular box with a thin black border, intended for sketching the poultry movement of the subject. The box occupies most of the page's width and height.