

# Indonesia

## Poster Presentation

# Poultry Production System (Layer and Broiler)

- **3 Main Challenges** to mitigate the risk of H5N1 virus at farmer's level
  - Limited regulation of poultry production throughout the country
  - General low standard of biosecurity on most farms
  - High density of sector 4 and high contact rate between sectors 3 and 4, both of which may contribute to H5N1 endemism in Indonesia
- **Lessons learned** in control to control/improve the risk of H5N1
  - Improved vaccination practices by farmers appears to have significantly reduced level of H5N1 virus in Indonesia
    - Use of local vaccine strains matched to circulating field virus
    - Farmers vaccinating during egg production rather than just during grow phase (pre-production)
  - Most occurrences of HN1 outbreaks in sector 4 (both poultry and ducks). Sector 4 may serve as the primary reservoir of the virus In Indonesia.
    - Passive surveillance by PDSR has been key in discovering the high level of circulation taking place in sector 4

# Actions to be undertaken in the next 5 years

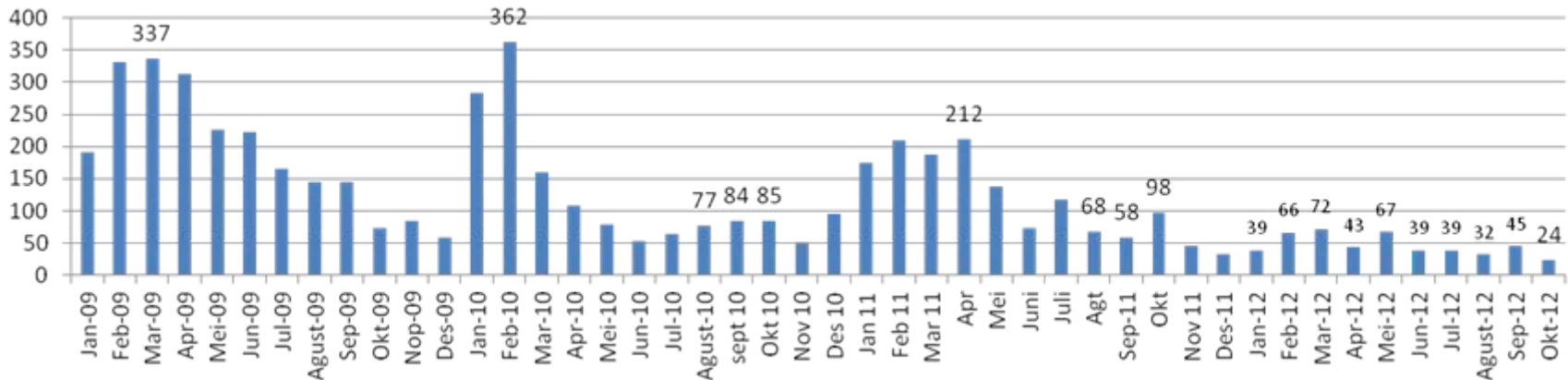
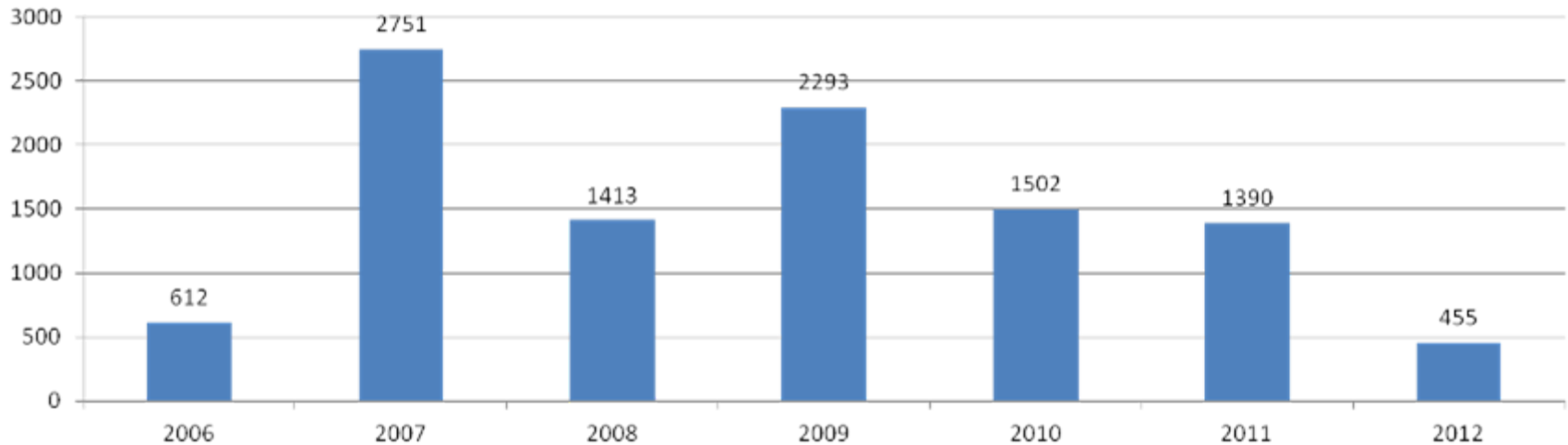
- In Indonesia, H5N1 virus is persistent and has been circulating in ecosystems and causing economic losses and public health concerns.
- Endemism appears to be driven by ongoing circulation in sector 4 (chickens and ducks) or combination of sectors 4 and 3.
- Greater effort is necessary to determine how to control virus in sector 4
- Lack of effective compensation is still an obstacle which decreases commercial farmer incentive for reporting and limits government's ability to contain large outbreak events
- Live bird markets in mega-urban areas appear a significant risk to spreading virus to humans, but not to poultry farms.

# Recent H5N1 Outbreaks in Domestic and Wild Birds

- Covering the following information .
  - (1) Number of outbreaks/cases
  - (2) Affected bird species (domestic and wild) birds
  - (3) Type of affected premises/locations (backyard, small scale farm, live bird markets, etc.)
  - (4) Geographical distribution of outbreaks
  - (5) Result of virus studies (subtype, clade, antigenic and genetic analysis, etc.
- **PDSR/SMS data**
- **DIC surveillance data**
- **All viruses within clade 2.1.3. Most within 2.1.3.2 subclade.**
- **No significant evolution within past 2 years.**

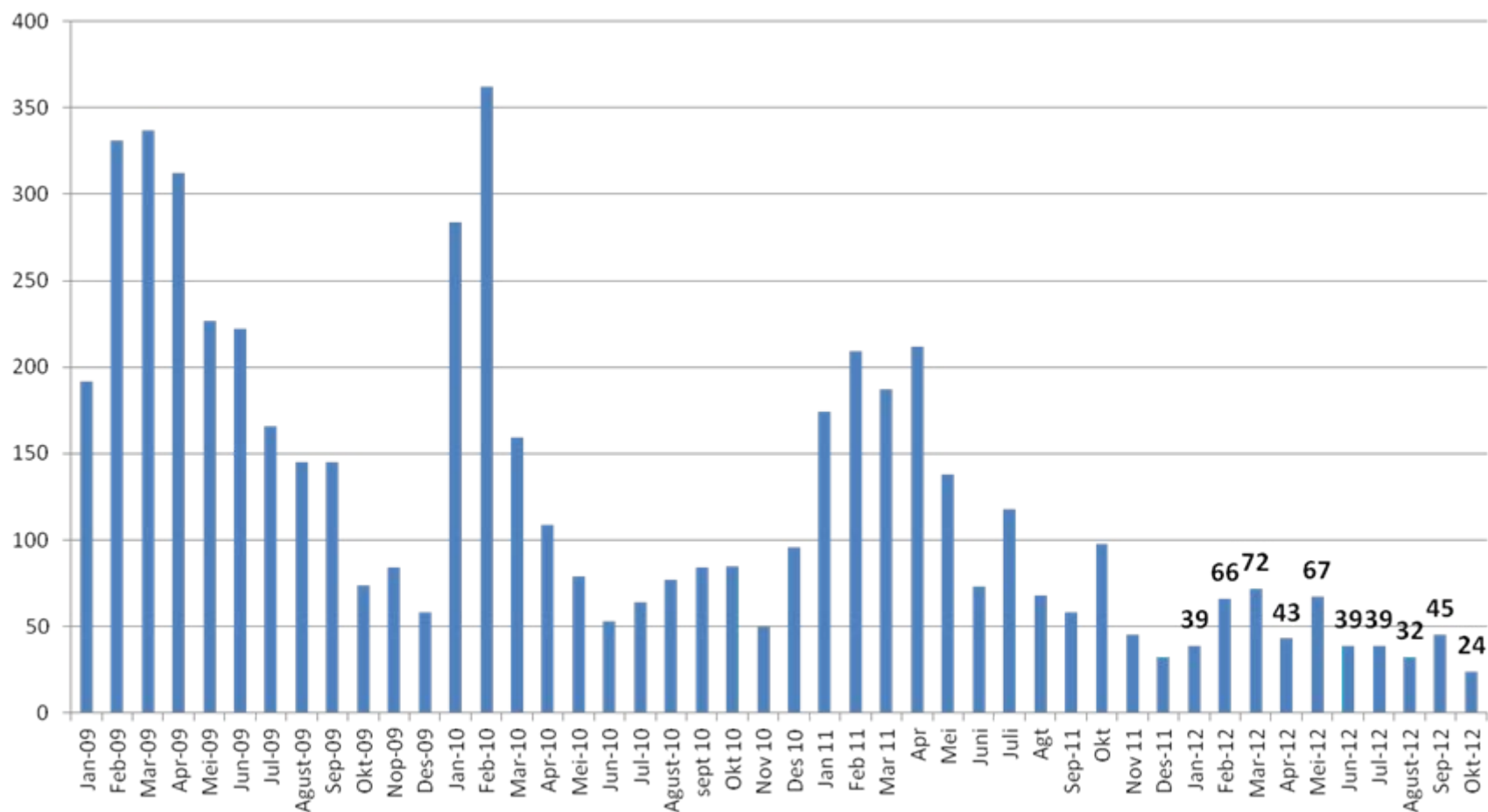


# Cases of HPAI in Backyard Poultry (mostly) by PDSR Data



# Geographical distribution of outbreaks HPAI

## January 2009 – October 2012



# Live Bird Markets

- Characteristics and main roles of Live bird markets are the primary means of retail sale of poultry throughout Indonesia
  - Sale of chilled or frozen poultry meat becoming more common in urban areas such as Jakarta.
- Over 10,000 live bird markets estimated in Indonesia
- Supply chain/distribution : Most live bird markets source poultry from wholesale “collector yards” which serve as an intermediate trading point between farm and market

# Surveillance Programme at poultry market chain

- A comprehensive programme was developed by DAH-FAO Indonesia to address surveillance and risk reduction along the post-production poultry market chain in the greater Jakarta area. Environmental surveillance of live bird markets and targeted surveillance of wholesale poultry collector yards has provided better information on the level of H5 virus circulating in the commercial industry, identified specific critical control points where risk of spread and amplification of H5 virus is high, and indicated the need for C&D interventions across the entire market chain to reduce the risk of H5 spread back to farms as well as to humans. Partnerships between poultry vendors, collector yard traders, and government have enabled the establishment of ongoing C&D activities and live bird market refurbishments across the greater Jakarta area.

- Government's intervention: Cleaning and disinfection programme implemented in live bird markets and collector yards in the greater Jakarta area since 2009.
  - Market cleaning days
  - Rehabilitation of sub-standard live bird markets
  - Truck cleaning stations at collector yards
  - Construction of consolidated collector yard/slaughterhouses with regulated SOPs for poultry slaughtering
  - Branding of safely-slaughtered whole poultry carcasses (Ayam ASUH) as means of increasing consumer awareness and demand for safer chilled carcasses rather than freshly slaughtered live birds.





Cleaning and disinfection of poultry crates at small collector yard



C&D station in operation



Cleaning and disinfection at slaughterhouse



Cleaning day training



**Cleaning day at Jakarta live bird market**



**Rehabilitation of live bird market**



**Before**



**After**

**Rehabilitation of outdoor live bird market**



**Before**



**After**



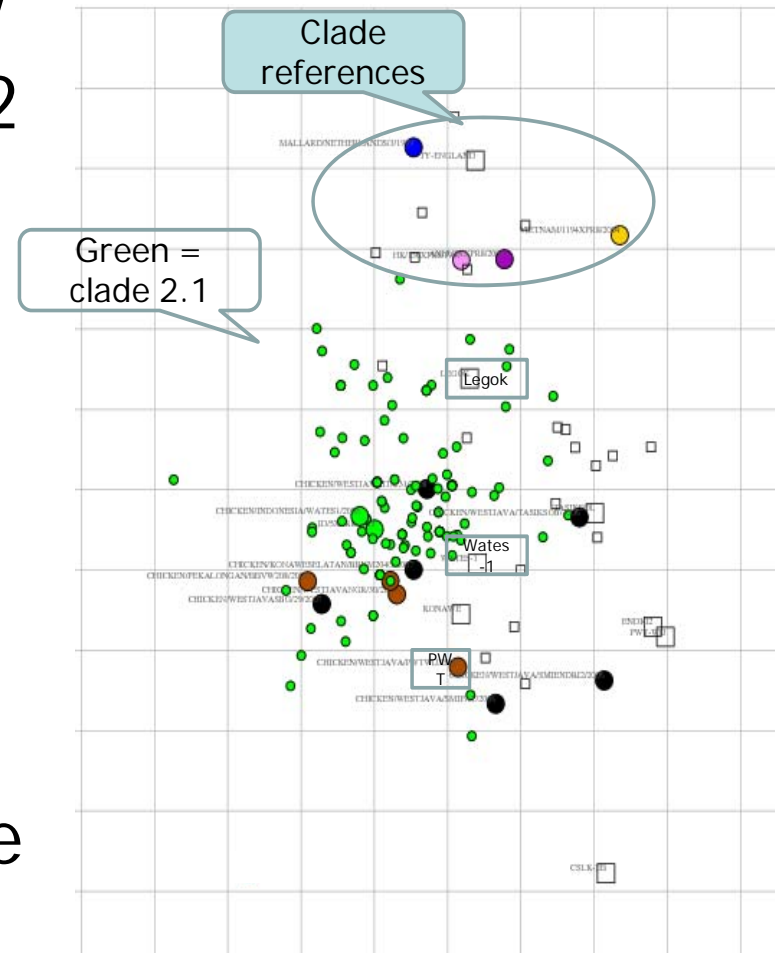
**Ayam ASUH for sale in Jakarta market**

- The realistic and feasible actions to be undertaken to improve the situation to reduce such risk of H5N1 at a live bird market level
- Due to the separation between farm and live bird markets, as well as the accumulation of virus contamination into downstream urban markets with limited contact with poultry farms, live bird markets do not appear to be the primary means of spread of the H5N1 virus to poultry, however live bird markets may be a primary means of spreading virus to human populations. There is no apparent relationship between endemic areas and locations with highly contaminated markets.



# Accomplished

- Identified 4 new antigenically relevant vaccine strains and 2 challenge strains for use by GOI and Indonesian vaccine manufacturers. New strain vaccines being produced.
- Pioneered application of antigenic cartography, a technique to characterize human influenza viruses since 2002, using avian sera.



# Active Surveillance Programme in Domestic Birds

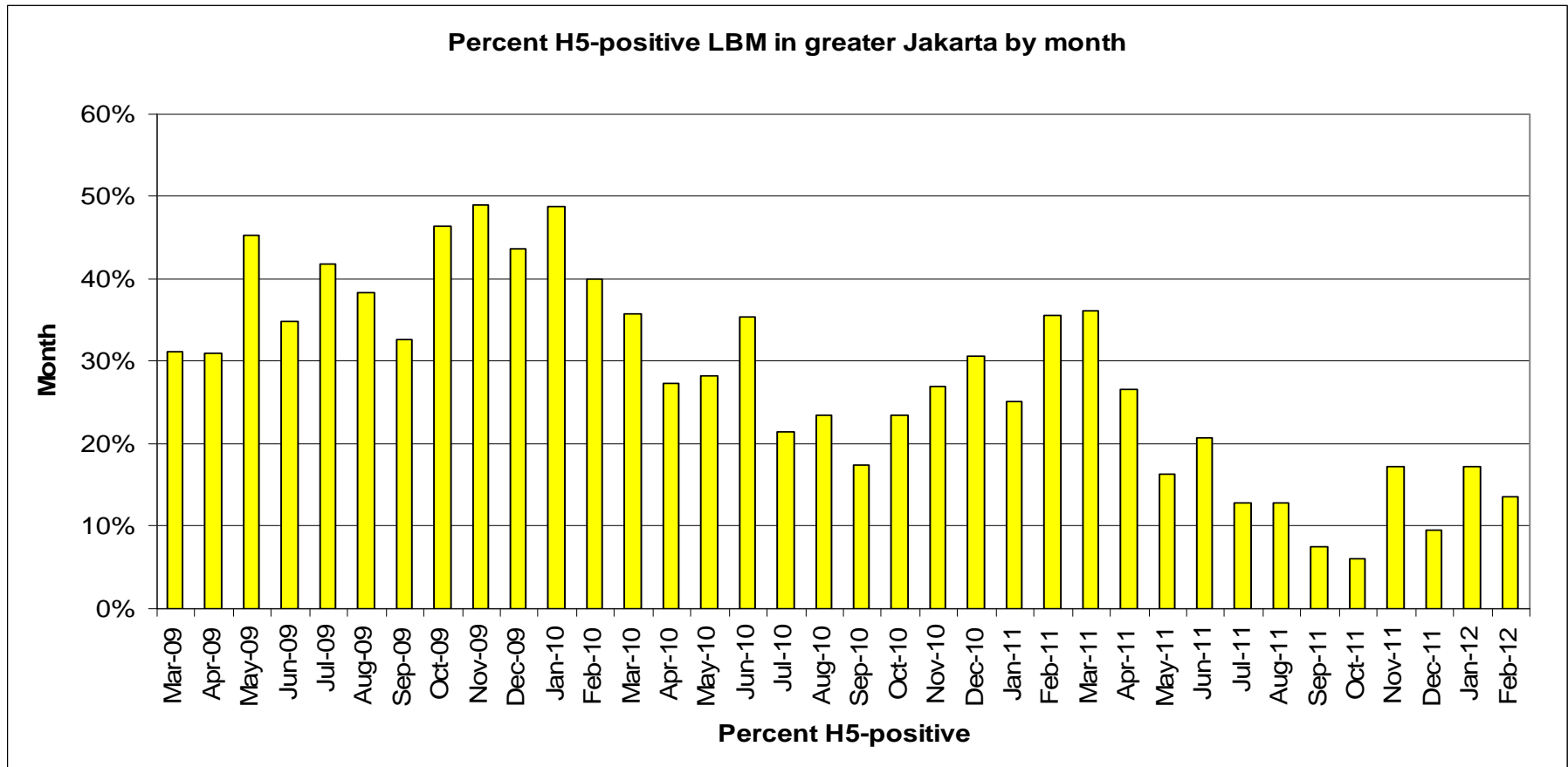
- In general, active surveillance is not a cost-effective means of detecting H5N1 virus in poultry
- In Indonesia, a specially designed live bird market surveillance programme is used to periodically determine the prevalence of H5-contaminated markets
  - The LBM environmental surveillance system is used to assess progress in controlling H5N1 in commercial poultry
- Active surveillance by local government is no longer supported because passive surveillance much more effective at detecting H5N1 outbreaks.

# Influenza A Virus Prevalence Studies in Indonesia

- Government of Indonesia and international organizations (FAO, IDP, etc) conducted influenza A virus prevalence studies on live poultry markets
- LBM Influenza A (matrix PCR) data
- Limited government financial and laboratory resources to study other influenza A viruses

# Live Poultry Market

1. Surveillance of prevalence in collector yard and traditional markets in Greater Jakarta. Declining trend of prevalence in commercial poultry farm since 2009-2012.



# Accomplishments of LBM surveillance

Goal: determine prevalence of H5-contaminated live bird markets over time.

Started in March 2009, 261 live bird markets included in the study. Environmental swabs from 5 sites pooled into one VTM sample. PCR testing for both matrix and H5

## Overall LBM surveillance results

From 5,862 environmental samples collected

**47.2% influenza A** virus positive

**26.2% H5** subtype positive results

The data showed a **progressively decreasing** prevalence in both matrix and H5-positive samples each year:

2009: **68.2%** matrix-positive and **40.1%** H5-positive

2010: **50.3%** matrix-positive and **28.8%** H5-positive

2011: **35.6%** matrix-positive and **18.9%** H5-positive

Seasonal pattern increasingly evident in 2010 and 2011

## Active/passive surveillance on wild birds, if any

- Due to the endemic nature of H5N1 in domestic poultry and limited control programme resources, active surveillance in wild birds is not a priority and not implemented.

# Country Presentation -India

Dr. A.S.Nanda

Animal Husbandry Commissioner  
Government of India



5<sup>th</sup> OIE Regional Expert Group Meeting for Implementation of the Programme on Surveillance  
of Wild and Domestic Birds along Migratory Flyways under the OIE/ Japan Trust Fund (JTF)  
Project for Strengthening HPAI Control in Asia  
Tokyo, Japan, 13-14 December, 2012

# HPNAI (H5N1) outbreaks (wef 2007)

Sl. No.	Period	State affected	Production system	Bird Type	Antigenic clade
1	July, 2007	Manipur	Farm	Chicken	2.2
2	Jan 2008	West Bengal	Back yard	Chicken	2.2
3	Apr, 2008	Tripura	Back yard	Chicken	2.2
4	Nov, 2008	Assam	Back yard	Chicken	2.2
5	Dec, 2008	West Bengal	Back yard	Chicken	2.2
6	Jan, 2009	Sikkim	Back yard	Chicken	2.2
7	Jan, 2010	West Bengal	Back yard	Chicken	2.2
8	Feb , 2011	Tripura	Farm	Duck	2.3.2.1
9	Sept, 2011	Assam	Back yard	Chicken	2.3.2.1



## HPNAI (H5N1) outbreaks (wef 2007); Contd.

Sl. No	Period	State affected	Production System	Bird Type	Antigenic clade
10	Sep., 2011	West Bengal	Back yard	Chicken	2.3.2.1
11	Jan., 2012	Odisha	Back yard	Chicken	2.3.2.1
12	Jan, 2012	Meghalaya	Farm	Chicken	2.3.2.1
13	Jan, 2012	Odisha	Back yard	Chicken	2.3.2.1
14	Jan, 2012	Tripura	Farm	Duck	2.3.2.1
15	Feb, 2012	Odisha	Farm	Chicken	2.3.2.1
16	Mar, 2012	Tripura	Farm	Chicken	2.3.2.1
17	Apr, 2012	Tripura	Farm	Chicken	2.3.2.1
18	Oct, 2012	Karnataka	Farm	Turkey	2.3.2.1

# Distribution of HPNAI outbreaks in domestic & wild birds



# AI Surveillance in Birds

## Two Types of Surveillance Programmes in Place:

### (1) Passive / Routine Surveillance :

- Routine observance for mortality
- Random sampling for virology
- Weekly reports (State Govts./ media reports/ industry/ universities/ diagnostic labs/ farmers/ public reports)

### (2) Active / Targeted Surveillance:

- Addresses unusual sickness/ mortality
- Daily reports received from the affected state

### AI Surveillance Involves:

(a) Physical /Clinical observance and reporting

(b) Laboratory Testing: Serum, cloacal and tracheal swabs in birds

# Active AI surveillance/ monitoring in domestic birds

## Three Stages of Active Surveillance:

(A) Pre-Confirmation (Suspected AI)

(B) Post- Operative Surveillance (After confirmed AI outbreak & culling)

(C) Post-Repopulation Surveillance (After induction of new birds)

## Major Surveillance Activities Include:

- Immediate physical confirmation of unusual sickness, mortality
- Collection and mailing of samples for laboratory diagnosis.

# **Active Surveillance: Collection of Samples & Dispatch**

## **Type of Samples & Mailing:**

- ❖ **Serum, cloacal swab, tracheal swab, feces, dead birds and morbid materials collected aseptically,**
- ❖ **Hand Carried, by Air (where possible) to OIE AI Reference Laboratory, Bhopal.**

## **Minimum Number of Samples to Confirm Outbreak**

- **5 recently dead or acutely sick, sacrificed birds for PME**
- **10 Serum samples from acutely sick birds**
- **10 Cloacal & tracheal swabs from apparently healthy birds**
- **5 sera samples from apparently healthy ducks, if present**

# **Active Surveillance: Samples Collection & Dispatch (Contd):**

**# Samples collected in a surveillance zone & tested at OIE lab.**

## **(B) Post- Operative Surveillance @ 15 days for 2 months**

- ❖ **Min. 2 birds in a unit of 1000 birds or 6 birds in larger units....OR**
- ❖ **Min. 6 birds in 50% of the villages in surveillance zone**

## **(C) Post-Repopulation Surveillance @ 15 days, for 2 months**

- ❖ **0.5% of population introduced (Or 2-6) in a farm or a village**

# Laboratory Methods Used for Testing AI

## (1) Serological tests:

- Agar Gel Immunodiffusion (AGID) and
- Haemagglutination Inhibition (HI)

## (2) Molecular Tests (RNA detection):

- Real time RT-PCR for matrix, H5 HA and N1 NA gene.
- RT-PCR for NP/matrix gene, H1 to H16 and N1 to N9 subtypes

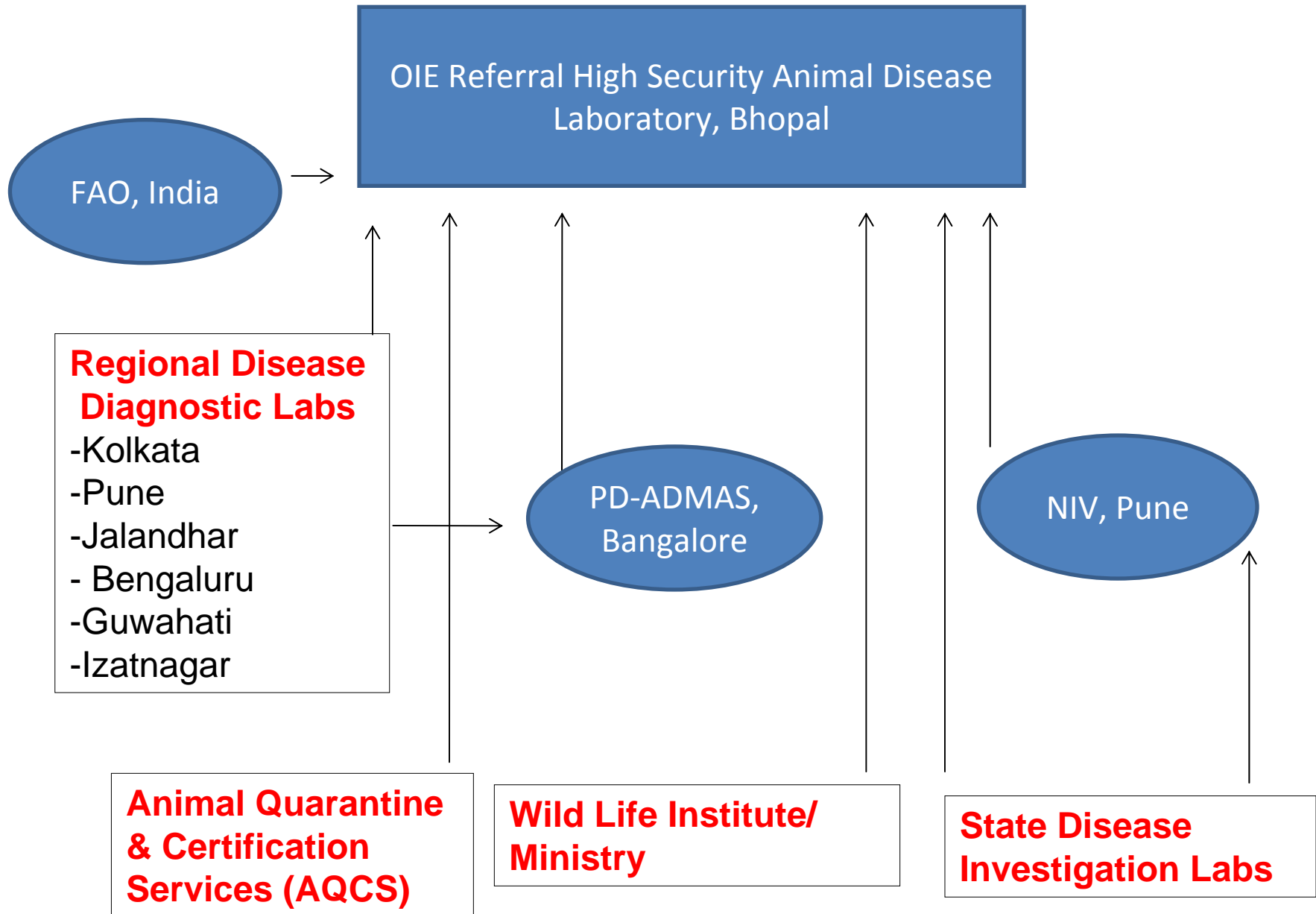
## (3) Virus isolation:

- In SPF embryonated chicken eggs

**Note: Action to contain AI is initiated on the receipt of positive NAI at the 1st stage of testing,**

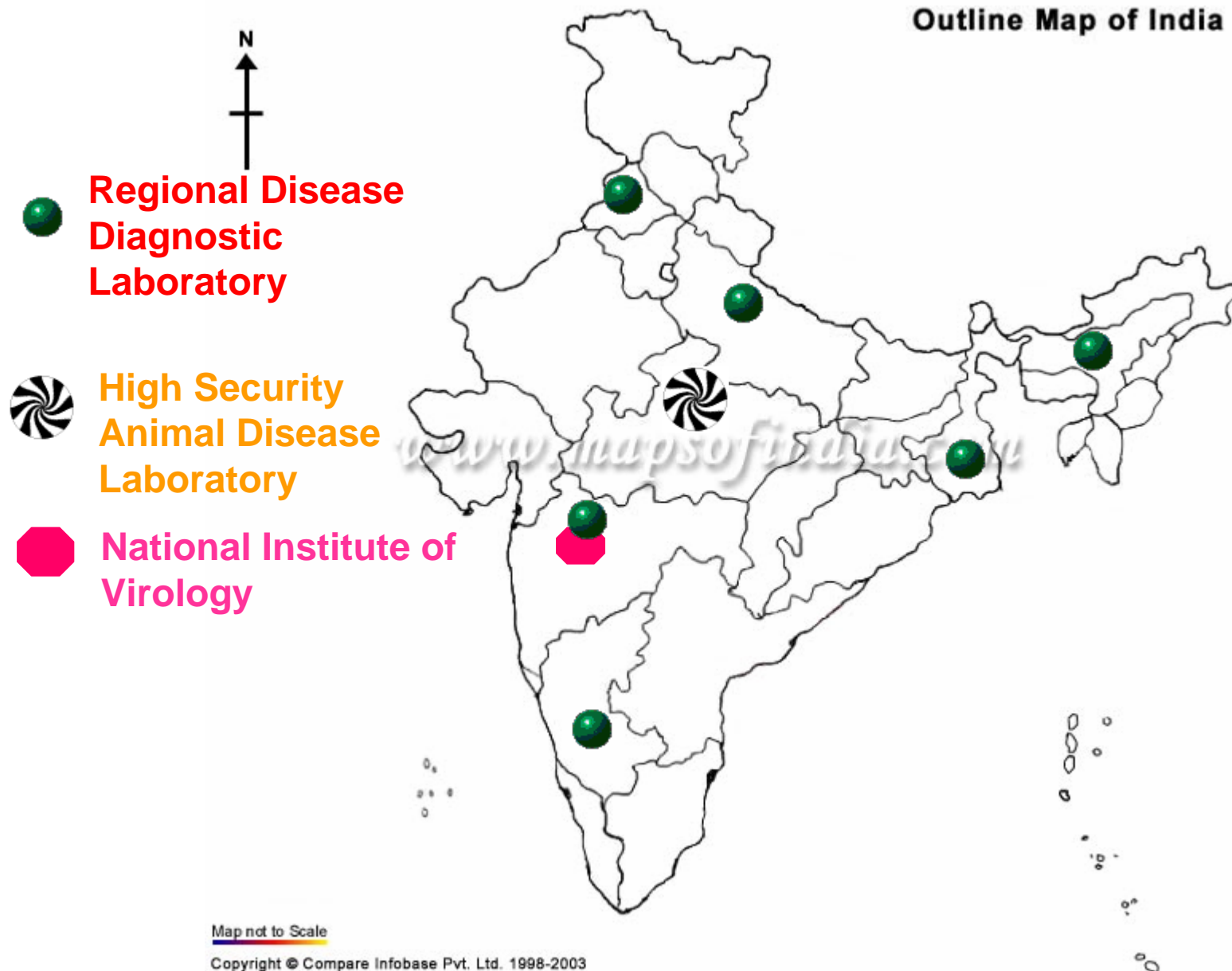
- **Since 2004, more than 700,000 samples have been tested for Influenza**

# Laboratory Network for Diagnosis of AI





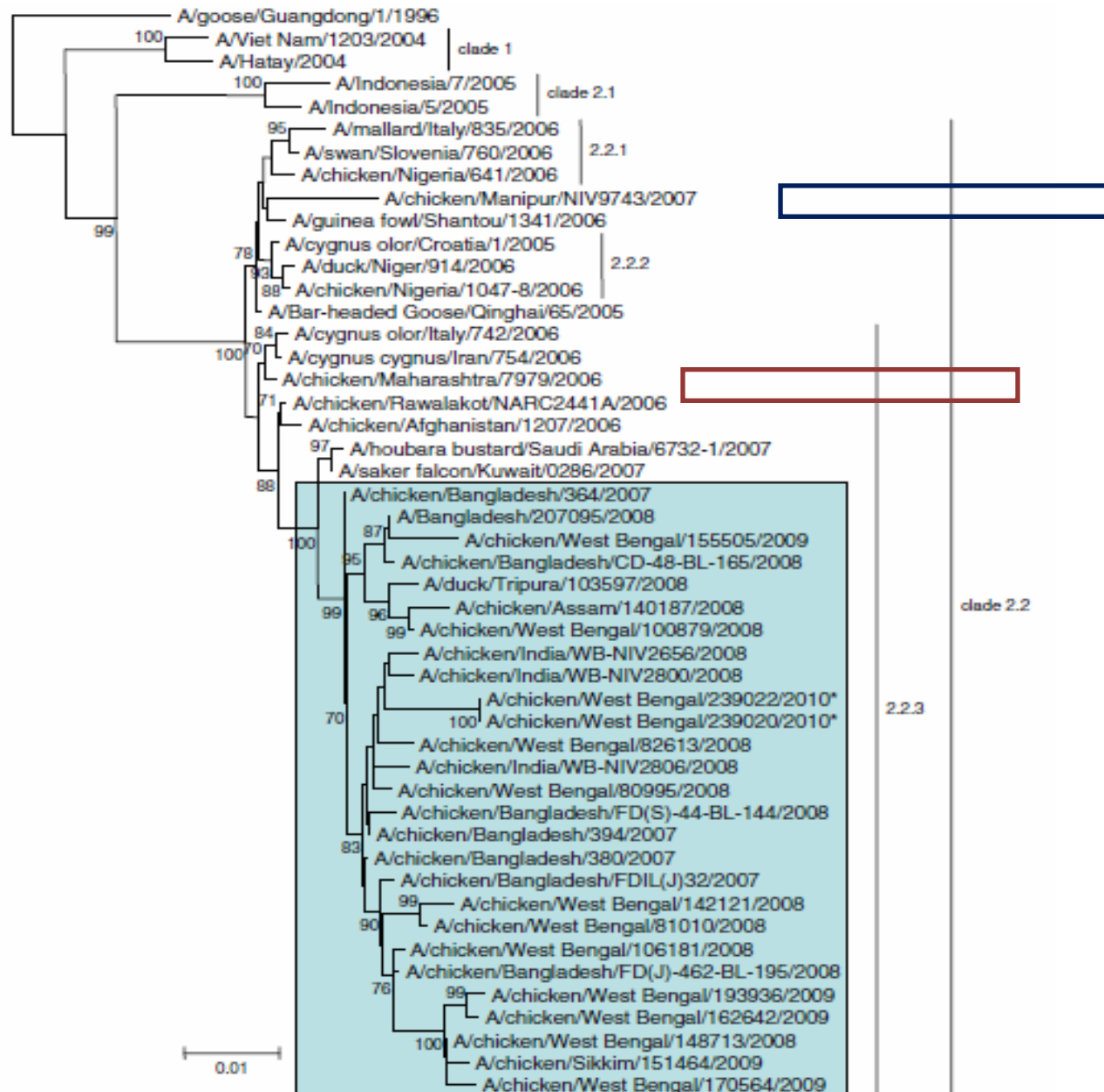
# Network of AI Surveillance laboratories



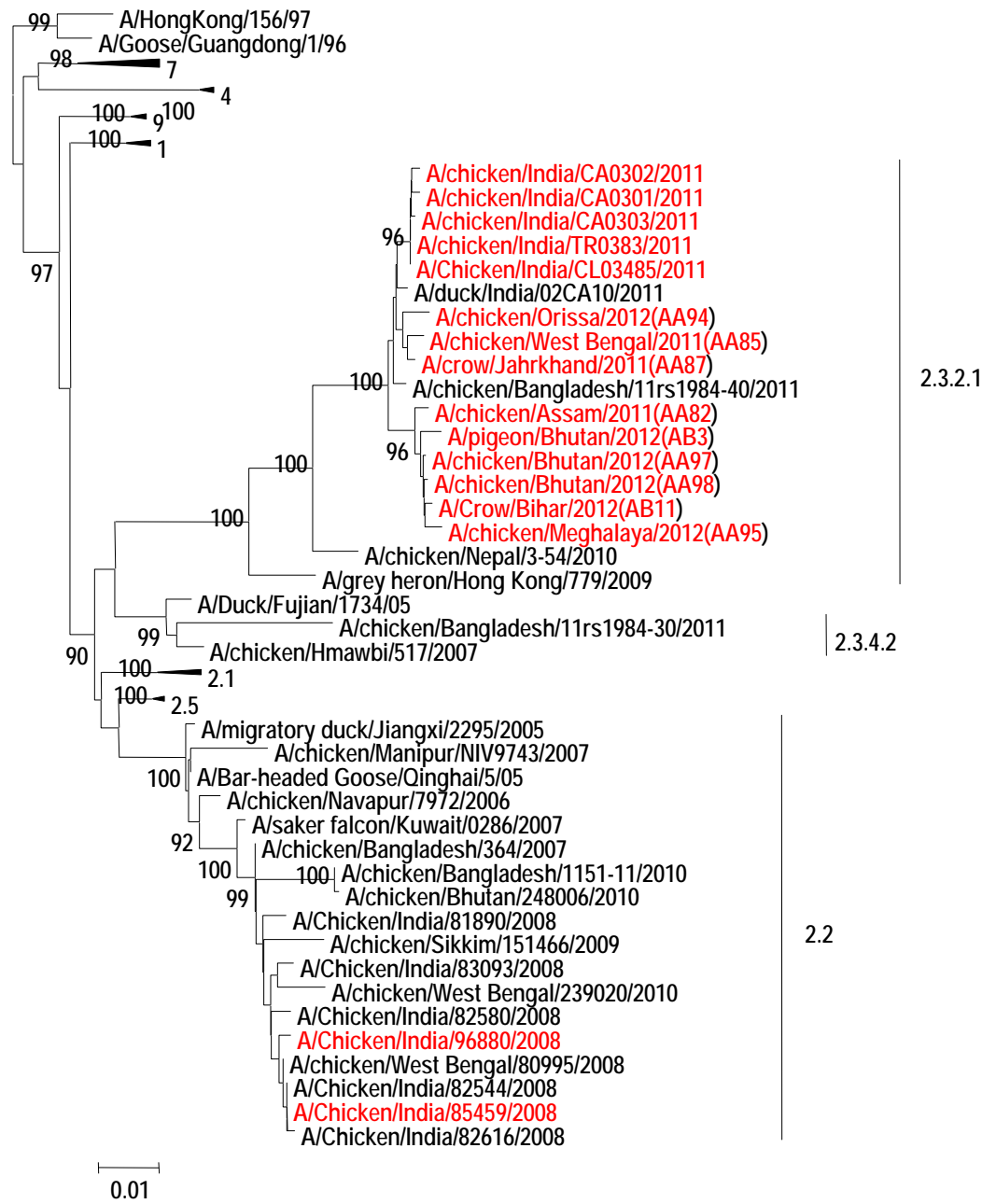
# **Molecular characteristics of Clade 2.2 H5N1 viruses**

- **Multiple basic amino acids RRRKKR\*GLF in all clade 2.2 viruses.**
- **Manipur virus sequence was RRRRKRR\*GLF.**
- **All had avian receptor ( $\alpha$ 2,3 sialic acid) binding specificity.**
- **A twenty-amino acid deletion in the NA stalk region (position 49-68), was similar to other genotype Z viruses and amino acid <sup>110</sup>R in NA gene unique for migratory birds.**
- **Oseltamivir resistance mutations <sup>E</sup>119<sup>A</sup> and <sup>N</sup>294<sup>S</sup> were found in some H5N1 viruses isolated from West Bengal in 2008.**
- **Some isolates had Lysine at position 627 of PB2 protein i.e. can infect mammals.**
- **S31N mutation in M2 ion channel protein only in 2010 viruses i.e. resistant to Amantadine.**

# Phylogenetic analysis of clade 2.2 H5N1 viruses



# Phylogenetic analysis of clade 2.3.2.1 H5N1 viruses

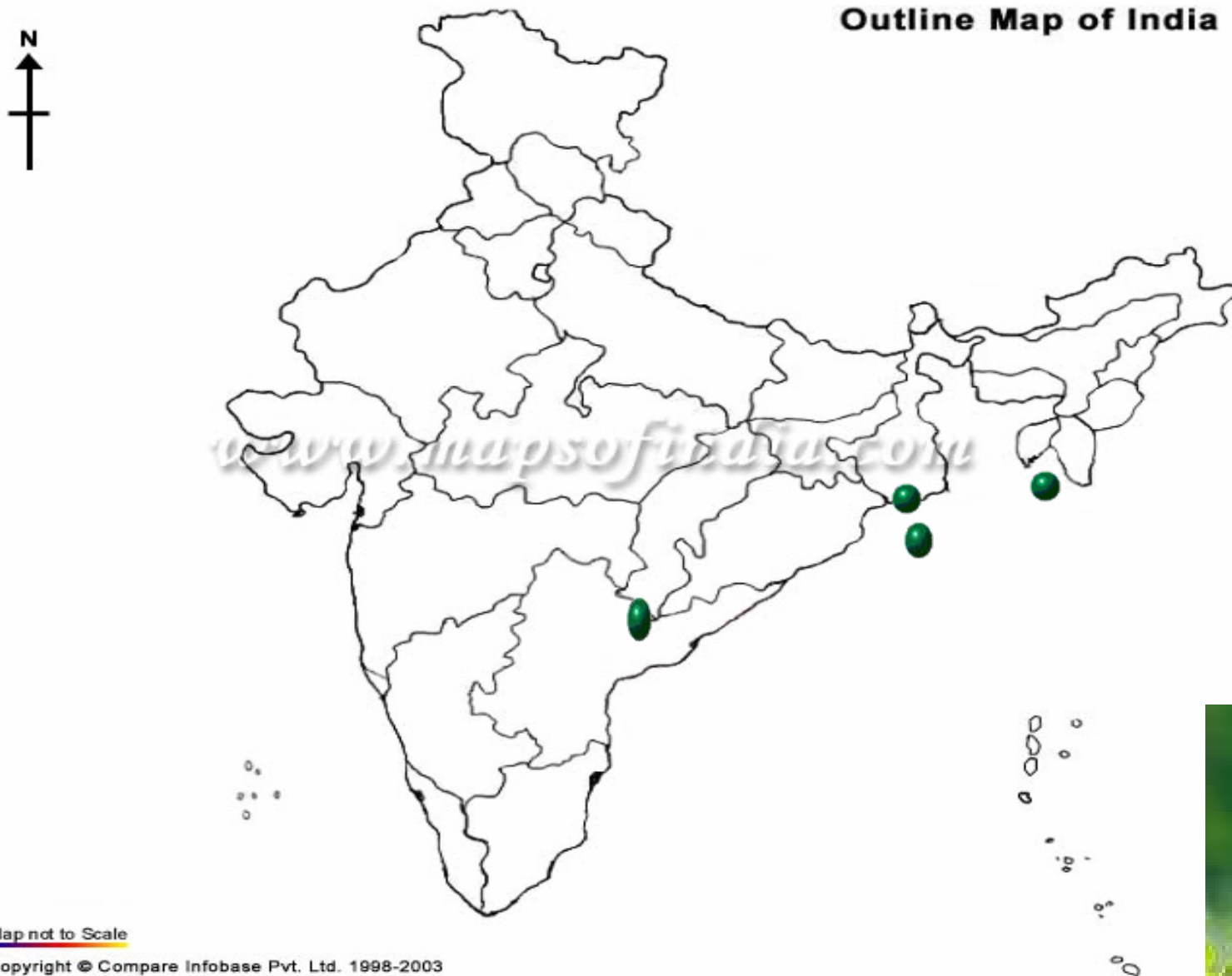


# AI Surveillance in Live Bird Markets (LBM)

- ~ 95 % of poultry meat supply is from Live Bird Markets.
- Birds mostly slaughtered/ auctioned/ sold there.
- NIV Pune monitors AI viruses in selected markets
- Few strains of non-notifiable AI viruses and some samples positive for H5N1 were recorded.
- Advisories Issued to concerned states to improve hygiene and to practice biosecurity to avoid transmission of AI.

# H5N1 in wild birds in India

Confirmed H5N1-HPAI in crows (2011-12)



# H5N1 reported in wild birds in India

- In 2011-12, some dead crows tested positive for H5N1
- Population of crows spread over a vast area involving several states
- Virus Clade = 2.3.2.1.

# AI Surveillance in Wild Birds in India

## Passive and Active AI surveillance done as in domestic birds

### **Material Used:**

- Dead birds
- Droppings from the area under surveillance
- Serum, tracheal and cloacal samples of randomly selected birds

### **Surveillance Plan:**

- Annual surveillance plan prepared and agreed to by October every year
- Surveillance Implemented by Veterinary staff and Wild life officials of concerned states
- FAO Officials also involved under USAID's 'Immediate Technical Assistance to Strengthen Emergency Preparedness for HPAI to India'

### **Test Facility:**

- HSADL, Bhopal
- NIV, Pune
- PD-ADMAS, Bangalore



# Implications of surveillance in wild birds

- Wild life broke routine bio-security barriers and might have helped spread of disease
- Wild birds traveled long distances that lead to faster spread of disease in vast areas
- Lead to better understanding of AI epidemiology, mutations, helped in forecasting and prevention of its further spread
- Declaration of the disease prompted suitable measures in place to contain it

# Some Important Observations

- Big backyard poultry sector with low biosecurity
- Unorganized Live bird markets – a big threat
- Surveillance played a big role in AI control
- Awareness campaigns have helped
- Legal back-up to containment programmes, and financial support for compensation was useful
- Regional collaboration (SAARC) is vital.

## Future Issues on AI & Plans-2013

- Continuance of the control and containment programme on AI
- Awareness programme and farmer participation in prompt reporting to be strengthened
- Strengthening of surveillance in live poultry markets
- Movement controls across International Borders
- Expand scope of epidemiological study
- Integrated Surveillance: **One Health** approach for animal, human and wild life sectors
- Strengthening of International as well as regional co-operation in control of AI

# The 5th OIE Regional Expert Group Meeting for Implementation of the Programme on Surveillance of Wild and Domestic Birds along Migratory Flyways under the OIE/Japan Trust Fund (JTF) Project for Strengthening HPAI Control in Asia

(Tokyo, Japan, 13-14 December 2012)



## Hong Kong SAR

**Dr. May Tse**  
**Veterinary Officer**

# Location of H5N1 positive cases found in Hong Kong (2007 – 2012)



**Year 2007**



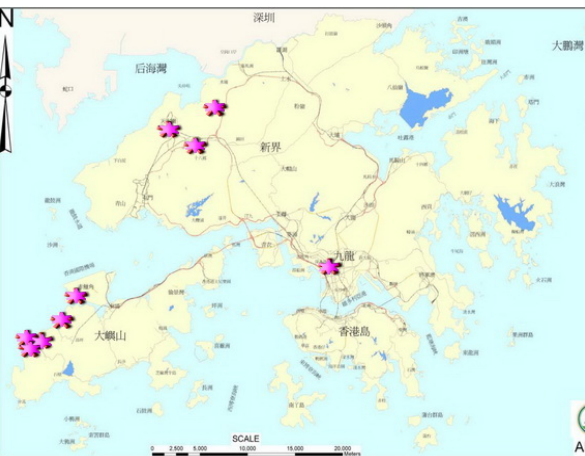
**Year 2008**



**Year 2009**



**Year 2010**



**Year 2011**



**Year 2012**  
up to 31 October 2012



# H5N1 infected birds found in Hong Kong (2007)



## Year 2007

Label 標号	Submission Date 樣本送交日期	Bird's name 雀鳥名稱	Quantity 數量	Location 地點
1	03.1.07	SCALY-BREASTED MUNIA 斑文鳥	1	29 LEIGHTON ROAD, WAN CHAI 灣仔禮頓道 29 號
2	12.1.07	CRESTED GOSHAWK 鳳頭鷹	1	NEAR SHEK KIP MEI HEALTH CENTRE, SHAM SHUI PO 深水步石硤尾健康院後山山頂
3	17.1.07	HOUSE CROW 家鴉	1	LAI ON ESTATE, SHAM SHUI PO 深水步麗安村
4	17.1.07	JAPANESE WHITE-EYE 相思	1	CONVAIR DRIVE, SAN PO KING 新蒲崗康維道
5	18.1.07	WHITE-RUMPED MUNIA 白腰文鳥	1	101-109 BOUNDARY STREET, MONGKOK 旺角界限街 101-109 號
6	24.1.07	PEREGRINE FALCON 遊隼	1	CHAI WAN KOK STREET, TSUEN WAN 荃灣柴灣角街致利工業大廈
7	25.1.07	HOUSE CROW 家鴉	1	YEE KOK COURT, SHAM SHUI PO 深水步怡閣苑怡康閣
8	07.2.07	BLUE MAGPIE 紅咀藍鵲	1	NORTH KOWLOON MAGISTRACY, TAI PO ROAD, SHAM SHUI PO 深水步大埔道北九龍裁判法院地下
9	09.2.07	SLIVER-EARED MESIA 銀耳相思鳥	2	101-109 BOUNDARY STREET, MONGKOK 旺角界限街 101-109 號
10	17.02.07	COMMON KESTREL 紅隼	1	SHING TIN HOUSE, PAK TIN ESTATE, SHAM SHUI PO 深水步白田邨盛田樓後垃圾站
11	21.02.07	CHESTNUT MUNIA 栗腹文鳥	1	near St Teresa's Hospital at Prince Edward Road West, Kowloon 太子道聖德肋撒醫院行人路
12	21.02.07	SCALY-BREASTED MUNIA 斑文鳥	1	Junction of Sing Woo Road and Wong Nai Chung Road, Happy Valley 跑馬地成和道黃泥涌交界
13	27.02.07	SCALY-BREASTED MUNIA 斑文鳥	1	Fu Yee House, Fu Cheong Estate, Sham Shui Po 深水步富昌邨富怡樓
14	06.03.07	Long-tailed Shrike 棕背伯勞	1	12 Hung Lok Road, Harbourview Horizon, Hung Hom 紅磡紅樂道 12 號海韻軒酒店
15	28.05.07	Starling (棕鳥), probably Red-billed Starling (相信為絲光棕鳥)	1	101-109 BOUNDARY STREET, MONGKOK 旺角界限街 101-109 號
16	04.06.07	COMMON MAGPIE 喜鵲	1	13 Jat Min Chuen St, Jat Min Chuen, Sha Tin 沙田乙明邨青年空間後門 (乙明邨街 13 號)
17	12.06.07	House Crow 家鴉	1	SHAM SHUI PO SPORTS GROUND 深水步運動場
18	20.11.07	Little Egret 小白鷺	1	Tuen Mun Park 屯門公園
19	5.12.07	Little Egret 小白鷺	1	Tuen Mun Park 屯門公園
20	6.12.07	Grey Heron 蒼鷺	1	MTR Lok Ma Chau Spurline wetland compensation area 港鐵落馬洲支線濕地彌償區
21	18.12.07	Common Buzzard 鵟	1	Sha Lo Wan, Lantau Island 大嶼山沙螺灣

Total number of positive cases = 21

# H5N1 infected birds found in Hong Kong (2008)

## Year 2008

Label 標号	Submission Date 樣本送交日期	Bird's name 雀鳥名稱	Quantity 數量	Location 地點
1	29.1.2008	BLACK-CROWNED NIGHT HERON 夜鷺	1	OCEAN PARK 海洋公園
2	04.02.2008	GREAT EGRET 大白鷺	1	TAM KON CHAU 擔竿洲路
3	09.02.2008	GREY HERON 蒼鷺	1	SAN SAM ROAD, LOK MA CHAU 落馬州新深路旁
4	11.02.2008	ORIENTAL MAGPIE ROBIN 鵲鴝	1	CHEUNG SHA WAN WHOLESALE FOOD MARKET 長沙灣副食品批發市場
5	03.03.2008	ORIENTAL MAGPIE ROBIN 鵲鴝	1	TAI PO KAU NATURE RESERVE 大埔滘自然護理區
6	10.03.2008	PEREGRINE FALCON, 遊隼	1	PARK ISLAND, MA WAN 馬灣珀麗灣
7	16.10.2008	HOUSE CROW 家鴉	1	SHAM SHUI PO PARK 深水埗公園

Total number of positive cases = **7**

# H5N1 infected birds found in Hong Kong (2009)



Year 2009

標号 Label	樣本送交日期 Submission Date	雀鳥名稱 Bird's Name	數量 Quantity	地點 Location
1	29.01.2009	鵝 goose	1	沙螺灣 Sha Lo Wan
2	29.01.2009	鴨 duck	1	沙螺灣 Sha Lo Wan
3	31.01.2009	鴨 duck	1	沙螺灣 Sha Lo Wan
4	02.02.2009	雞 chicken	1	沙螺灣 Sha Lo Wan
5	02.02.2009	雞 chicken	1	深屈灣 Sham Wat Wan
6	02.02.2009	鴨 duck	1	深屈灣 Sham Wat Wan
7	03.02.2009	雞 chicken	1	屯門蝴蝶灣泳灘 Butterfly Beach, Tuen Mun
8	03.02.2009	鴨 duck	1	磡石灣 San Shek Wan
9	04.02.2009	雞 chicken	1	大澳新基街 Sun Ki Street, Tai O
10	02.02.2009	蒼鷺 Grey Heron	1	米埔自然保護區 Mai Po Nature Reserve
11	03.02.2009	游隼 Peregrine Falcon	1	油柑頭 Yau Kam Tau
12	05.02.2009	大咀烏鴉 Large-billed Crow	1	駿群路古窯公園 Chun Kwan Road Ancient Kiln Park
13	12.02.2009	八哥 Crested Myna	1	平洲 (又名東平洲) 沙頭 Sha Tau, Ping Chau (aka Tung Ping Chau)
14	03.03.2009	雞 Chicken	1	平洲 (又名東平洲) 更樓石營地 Kang Lau Shek, Ping Chau (aka Tung Ping Chau)
15	27.04.2009	鴿 Feral Pigeon	1	屯門良德街 Leung Tak Street, Tuen Mun
16	30.12.2009	鵲鴝 Magpie Robin	1	鶴藪管理站 Hok Tau management centre

Total number of positive cases = **16**



# H5N1 infected birds found in Hong Kong (2010 & 2011)



## Year 2010

標号 Label	樣本送交日期 Submission Date	雀鳥名稱 Bird's Name	數量 Quantity	地點 Location
1	29 March, 2010	家燕 Barn Swallow	1	米埔新村 Mai Po San Tsuen
2	20 December, 2010	雞 Chicken	1	大嶼山沙螺灣 Sha Lo Wan, Lantau Island

Total number of positive cases = **2**

## Year 2011

標号 Label	樣本送交日期 Submission Date	雀鳥名稱 Bird's Name	數量 Quantity	地點 Location
1	18 January, 2011	鵲鴝 Oriental Magpie Robin	1	元朗錦繡花園 Fairview Park, Yuen Long
2	19 January, 2011	大嘴烏鴉 Large Billed Crow	1	大澳新基街 Sun Ki Street, Tai O
3	25 January, 2011	紅嘴鷗 Black-headed Gull	1	大嶼山南環路81號 81 South Perimeter Road, Lantau
4	25 January, 2011	雞 Chicken	1	大澳楊侯古廟 附近 Near Yeung Hau Temple, Tai O
5	28 January, 2011	雞 Chicken	2	大澳楊侯古廟寶珠潭附近的海灘 Beach near Po Chue Tam, Yeung Hau Temple, Tai O
6	4 February, 2011	鴨 Duck	1	大澳楊侯古廟寶珠潭附近的海灘 Beach near Po Chue Tam, Yeung Hau Temple, Tai O
7	1 March, 2011	鵝 Goose	1	大嶼山深石村附近的海灘 Beach near Sham Shek Tsuen, Lantau
8	14 December, 2011	紅嘴鷗 Black-headed Gull	1	元朗體育路7號趙聿修紀念中學 Chiu Lut Sau Memorial Secondary School, 7 Tai Yuk Road, Yuen Long
9	19 December, 2011	鵲鴝 Oriental Magpie Robin	1	天水圍天湖路5號裘錦秋中學 (元朗) Ju Ching Chu Secondary School (Yuen Long), 5 Tin Wu Road, Tin Shui Wai
10	20 December, 2011	雞 Chicken	1	長沙灣臨時家禽批發市場 Cheung Sha Wan Temporary Wholesale Poultry Market

Total number of positive cases = **10**

# H5N1 infected birds found in Hong Kong (2012)



## Year 2012

標号 Label	樣本送交日期 Submission Date	確診日期 Date of Confirmation	雀鳥名稱 Bird's Name	數量 Quantity	地點 Location
1	2 January, 2012	6 January, 2012	Black-headed Gull 紅嘴鵠	1	屯門龍門路133號環保園 EcoPark, 133 Lung Mun Road, Tuen Mun
2	2 January, 2012	6 January, 2012	Black-headed Gull 紅嘴鵠	1	大嶼山沙螺灣足球場附近的渠道 A drain near Sha Lo Wan Soccer Pitch, Lantau
3	13 January, 2012	17 January, 2012	Little Egret 小白鷺	1	元朗南生圍第123約地段第1520號餘段漁護署濕地及動物護理(執法)組元朗辦事處前 Outside Yuen Long Office, Wetland and Fauna Conservation (Enforcement) Section of APCD, Lot 1520RP, DD123, Nam Sang Wai, Yuen Long
4	20 January, 2012	26 January, 2012	Goose 鵞	1	深井青山公路13哩釣魚灣泳灘 Anglers's Beach, 13 milestone, Castle Peak Road, Sham Tseng
5	31 January, 2012	3 February, 2012	Peregrine Falcon 遊隼	1	東涌文東路海堤灣畔的公園 The park of Seaview Crescent, Man Tung Road, Tung Chung
6	31 January, 2012	3 February, 2012	Black-headed Gull 紅嘴鵠	1	元朗後賢坊遊樂場 Chun Yin Square Playground, Yuen Long
7	31 January, 2012	6 February, 2012	Oriental Magpie Robin 鸚鵡	1	屯門友愛邨順德聯誼總會何日東小學 Shun Tak Fraternal Association Ho Yat Tung Primary School, Yau Oi Estate, Tuen Mun
8	7 February, 2012	13 February, 2012	Little Egret 小白鷺	1	屯門鄉事會路屯門公園北面入口 The northern entrance to Tuen Mun Park, Tuen Mun Heung Sze Wui Road, Tuen Mun
9	8 February, 2012	13 February, 2012	Little Egret 小白鷺	1	屯門鄉事會路屯門公園北面入口 The northern entrance to Tuen Mun Park, Tuen Mun Heung Sze Wui Road, Tuen Mun
10	8 February, 2012	14 February, 2012	Oriental Magpie Robin 鸚鵡	1	元朗橫瀾東頭圍新村2巷14號天台 The rooftop of No 14, Lane 2, Tung Tau Wai San Tsuen, Wang Chau, Yuen Long
11	9 February, 2012	14 February, 2012	Grey Heron 蒼鷺	1	元朗南生圍路山貝河附近的草地 The grassland near Shan Pui River, Nam Sang Wai Road, Yuen Long
12	27 February, 2012	5 March, 2012	Crested Goshawk 鳳頭鷹	1	旺角亞答街131號拔萃男書院 Diocesan Boys' School, 131 Argyle Street, Mong Kok
13	27 February, 2012	5 March, 2012	Oriental Magpie Robin 鸚鵡	1	長洲興隆後街76號3樓天台 The rooftop, 3/F, 76 Hing Lung Back Street, Cheung Chau
14	28 February, 2012	5 March, 2012	Oriental Magpie Robin 鸚鵡	1	長洲學佬巷80號 80 Hok Loo Lane, Cheung Chau
15	5 March, 2012	12 March, 2012	Black-headed Gull 紅嘴鵠	1	屯門青山發電廠B廠 Castle Peak Power Station Plant B, Tuen Mun
16	5 March, 2012	12 March, 2012	Peregrine Falcon 遊隼	1	馬鞍山錦豐苑錦葵閣 Kam Kwai House, Kam Fung Court, Ma On Shan
17	13 March, 2012	19 March, 2012	Peregrine Falcon 遊隼	1	大嶼山南環路80號 80 South Perimeter Road, Lantau
18	16 March, 2012	22 March, 2012	House Crow 家鴉	1	石硤尾大坑東遊樂場外一個油站旁的花園 A planter next to a petrol filling station, outside Tai Hang Tung Recreation Ground, Shek Kip Mei
19	5 April, 2012	10 April, 2012	Oriental Magpie Robin 鸚鵡	1	紅磡差館里十九號附近 Near 19 Station Lane, Hung Hom
20	30 April, 2012	7 May, 2012	Crested Myna 八哥	1	元朗加州豪園蒙尼卡徑附近 Near Santa Monica Avenue, Royal Palms, Yuen Long
21	19 June, 2012	25 June, 2012	Scaly-breasted Munia 斑文鳥	1	元朗康樂街28號金巴崙長老會耀道中學 Cumberland Presbyterian Church Yao Dao Secondary School, 28 Hong Yip Street, Yuen Long
22	1 July, 2012	5 July, 2012	House Crow 家鴉	1	九龍塘又一村丹桂路10號 10 Osmanthus Road, Yau Yat Chuen, Kowloon Tong

Total number of positive cases = **22**

# HPAI surveillance in Hong Kong SAR



## Test Methods (for both domestic and wild birds)

- Necropsy & histopathology examination
- Immunoperoxidase test on frozen tissue sections
- Virus isolation
- AI specific genome detection by PCR methods
- Anti-H5 antibody detection (i.e. Haemagglutination Inhibition test- HI test)



# HPAI surveillance in Hong Kong SAR



	<u>Location</u>	<u>Avian Type</u>	<u>Sample Type</u>
<b>Domestic birds</b>	Local Farm	Poultry (live, dead)	Clotted blood, Tracheal and cloacal swabs, Tissue, Environmental swab
	Import at MKT	Poultry (live, dead)	Clotted Blood, Tracheal and cloacal swabs, Tissue, Frozen or chilled poultry product swab
	Retail Poultry Market	Poultry (live, dead)	Tracheal and cloacal swabs, Tissue, Environmental swab
	Wholesale Poultry Market	Poultry (live, dead)	Clotted blood Tracheal and cloacal swabs, Tissue, Environmental swab
	Other locations (eg. Backyard, dead wild poultry, TLVL etc.)	Poultry (live, dead & stray)	Clotted blood, Tracheal and cloacal swabs, Tissue, Environmental swab
	Pet bird shop	Bird (live)	Environmental swab, Faecal dropping
<b>Wild birds</b>	Wild bird collection (eg. Urban area, country parks, wetland park etc.)	Bird (live, dead)	Tracheal and cloacal swabs, Tissue, Faecal swab

# Active surveillance program on **domestic birds** in Hong Kong

## Import Control

- 70%(14/20) blood samples collected from every consignment (2000 birds), H5 HI titre level  $> 1/16$  .
- Cloacal swabs for PCR testing.
- Quarantine Detector Dogs patrol at the borders and airport.
- Cloacal swabs from illegal birds are submitted to Vet Lab for PCR testing.

## Local poultry wholesale market

- Daily check for dead birds.
- Cloacal swabs for PCR / virus isolation.
- Random faecal/cage/floor swabs for virus isolation. (30 pool samples /month from CSWTPWM)

## Surveillance in pet bird market

- Regular swabs (faecal or cage) are submitted for lab testing.

## Local chicken farms

- Bird-proof nets, control movements of birds, humans and equipment, vaccination, disinfection, record,... etc.
- Farm visit, monthly bird count, dead birds sampling, presale sample for testing and issue of approval code, etc.
- Sentinel birds placed in vaccinated flocks to detect incursion of AIVs.
- Sentinel health condition/head count and H5 HI titer as criteria for market sale.
- Random faecal/cage/floor swabs for virus isolation monthly.



# Active/Passive surveillance program on **domestic birds** in Hong Kong



Year	No. of positive H5N1 case					
	Dead Wild Poultry	Wholesale Mkt.	Retail Mkt.	Local Farm	Park	Pet bird
2007	0	0	0	0	0	1
2008	0	0	4	3	0	0
2009	10	0	0	0	0	0
2010	1	0	0	0	0	0
2011	5	1	0	0	0	0
2012(up to Oct)	1	0	0	0	0	1
Total	17	1	4	3	0	2

# Active/Passive surveillance program on **domestic birds** in Hong Kong



## AI Antibody Testing For Imported Poultry Control Statistics 2007-2009

	2007		2008		2009	
	Number of blood sample for Ab testing	Percentage	Number of blood sample for Ab testing	Percentage	Number of blood sample for Ab testing	Percentage
Local Poultry Farm	58703	39.75%	42770	39.62%	72344	61.77%
Import Poultry	88966	60.25%	65184	60.38%	41916	35.79%
Poultry Market	0	0%	0	0%	2534	2.16%
Park bird	0	0%	0	0%	330	0.28%
<b>Total</b>	<b>147669</b>	<b>100.00%</b>	<b>107954</b>	<b>100.00%</b>	<b>117124</b>	<b>100.00%</b>



# Active/Passive surveillance program on **domestic birds** in Hong Kong



## AI Antibody Testing For Imported Poultry Control Statistics 2010-2012 (Oct)

	2010		2011		2012 (up to 31 Oct)	
	Number of blood sample for Ab testing	Percentage	Number of blood sample for Ab testing	Percentage	Number of blood sample for Ab testing	Percentage
Local Poultry Farm	85140	62.53%	74558	61.31%	68849	63.51%
Import Poultry	46370	34.06%	42680	35.09%	35732	32.96%
Poultry Market	4446	3.27%	4202	3.46%	3645	3.36%
Park bird	195	0.14%	178	0.15%	182	0.17%
<b>Total</b>	<b>136151</b>	<b>100.00%</b>	<b>121618</b>	<b>100.00%</b>	<b>108408</b>	<b>100.00%</b>



# Active/Passive surveillance program on **wild birds** in Hong Kong

## ➤ Active surveillance

- AI Monitoring and surveillance of wild bird droppings from wetland areas
- Since the end of 2002, the surveillance programme has been extended to cover wild birds, captive wild birds in recreational parks and pet birds in retail bird markets.

## ➤ Passive surveillance

- Passive surveillance is more sensitive than active surveillance in wild birds in detecting HPAI.
- Twenty-four hours sick and dead birds collection service for AI testing starting from 27 October 2005.



# Active/Passive surveillance program on **wild birds** in Hong Kong



	Year					
	2007	2008	2009	2010	2011	2012 (up to Oct)
Number of sample tested	15,519	13,940	15,238	11,815	11,760	14,872
Number of bird infected with H5N1 virus	21	7	6	1	5	21

# Summary of AI control and measures against HPAI



<i>Suspected Case</i>	Criteria	Corresponding Response Level	AFCD Actions
<b>Local Farm</b>	Positive H5 AI environmental swab <b>OR</b> Sentinel chicken with positive serology for H5 AI	Alert Response Level	<ul style="list-style-type: none"> <li>• Farm placed under quarantine</li> <li>• Suspension of sales</li> <li>• Intensive testing of whole farm</li> <li>• Virus isolation and characterization</li> <li>• Increased surveillance of farms</li> </ul>
<b>Wholesale Market</b>	Positive H5 AI environmental swab	Alert Response Level	<ul style="list-style-type: none"> <li>• Tracing back to farm origin</li> <li>• Intensive testing of farm if local farm origin</li> </ul>
<b>Retail Market</b>	Positive H5 AI environmental swab	Alert Response Level	<ul style="list-style-type: none"> <li>• Tracing back to farm origin</li> <li>• Intensive testing of farm if local farm origin</li> </ul>
<b>Pet Bird Shop</b>	Positive H5 AI environmental swab <b>OR</b> Dead pet bird with PCR positive H5 AI	Alert Response Level	<ul style="list-style-type: none"> <li>• Shop placed under quarantine</li> <li>• Suspension of sales</li> <li>• Intensive testing of shop stock</li> </ul>
<b>Imported Pet Bird</b>	Serological or PCR positive H5 AI imported pet bird in quarantine	Alert Response Level	<ul style="list-style-type: none"> <li>• Suspension of imports from country of origin</li> <li>• Virus isolation and characterization</li> </ul>
<b>Wild Bird</b>	Dead Wild bird with PCR positive H5 AI	Alert Response Level	<ul style="list-style-type: none"> <li>• Increase surveillance of wild birds</li> <li>• Increase surveillance on local poultry farms</li> <li>• Virus isolation and characterization</li> </ul>
<b>Bird in recreational Park</b>	Dead bird in recreational park with PCR positive H5 AI	Alert Response Level	<ul style="list-style-type: none"> <li>• Park placed under quarantine</li> <li>• Intensive testing of park birds</li> <li>• Virus isolation and characterization</li> </ul>

# Summary of AI control and measures against HPAI



<i>Confirmed Case</i>	<b>Criteria</b>	<b>Corresponding Response Level</b>	<b>AFCD Actions</b>
<i>Imported Pet Bird</i>	Confirmed case by virus isolation or PCR in a pet bird held in quarantine	Alert Response Level	<ul style="list-style-type: none"> <li>• Depopulate all birds in quarantine</li> </ul>
<i>Wild Bird</i>	Confirmed case by virus isolation or PCR in a wild bird	Alert Response Level	<ul style="list-style-type: none"> <li>• Increase surveillance and testing of wild birds</li> <li>• Increase surveillance and monitoring on local poultry farms</li> <li>• Closure of Wetland Park and Mai Po Nature Reserve and other walk-in aviaries if dead wild birds are found within 3-km radius of the parks, premises or facilities.</li> </ul>
<i>Bird in Recreational Park</i>	Confirmed case by virus isolation or PCR in a bird in a recreational park	Alert Response Level	<ul style="list-style-type: none"> <li>• Close and quarantine recreational parks</li> </ul>
<i>Pet Bird Shop</i>	Confirmed case by virus isolation or PCR in a bird in a pet bird shop	Alert Response Level	<ul style="list-style-type: none"> <li>• Quarantine and closure of shop plus any adjacent pet bird shops.</li> <li>• Depopulation of the pet bird shop as necessary</li> </ul>
<i>Local Farms, Wholesale or Retail Markets</i>	Any suspected case in local farms, wholesale or retail markets confirmed by virus isolation or PCR with no evidence of farm to farm spread	Serious Response Level	Depopulation of infected premises and quarantine and surveillance of neighbouring farms
<i>Outbreak in Local Farms, Wholesale or Retail Markets</i>	Any suspected outbreak in local farms, wholesale or retail markets confirmed by virus isolation or PCR with evidence of farm to farm spread	Serious Response Level	Depopulation of all premises and farms

# Actions to be undertaken in Asia in the next 5 years



## 1. Increase AI surveillance and monitoring and sharing the information

*- To understand, assess and monitor the HPAI situation and its spread*

## 2. Analyze the most recent circulating viruses in the regional and vaccine matching results and sharing the information

*- To assess and monitor the effectiveness of AI vaccination and vaccines used*

## 3. Enhance the implementation of control measures such as AI vaccination and stamping out policy

*- To prevent and/or reduce the spread of HPAI to other areas/countries in the region*



**Thank you**

**The 5th OIE Regional Expert Group Meeting for Implementation of the Programme on  
Surveillance of Wild and Domestic Birds along Migratory Flyways  
under the OIE/Japan Trust Fund (JTF) Project for Strengthening HPAI Control in  
Asia**

# **AIV Diagnosis and Surveillance**

National/OIE AI Reference Laboratory,  
Harbin Veterinary Research Institute, China

# **Report-1: Recent HPAI outbreaks and H5N1 HPAI virus characterization in domestic and wild birds**



# HPAI outbreaks occurred in P. R. China (from 2007)

Time of outbreaks	Subtype	Clade	Province /City	No.Death(D)/Sick(S)	Affected bird species	Type of affected premises/locations
2012.9	H5	2.3.2	Guangdong/Zhanjiang	6300/14050	Duck	Backyard
2012.6	H5	2.3.2	Xinjiang	1600/5500	Chicken	Backyard
2012.6	H5	7.2	Gansu/Baiyin	260/6200	Chicken	Backyard
2012.4	H5	2.3.2	Liaoning/Dalian	5/5	Chicken	Kindergarten
2012.4	H5	7.2	Ningxia/Guyuan	23880(S)	Chicken	Backyard
2011.12	H5	2.3.2	Tibet/Lasa	290(D)	Chicken	Backyard
2010.6	H5	2.3.2	Tibet/Naqu	170(D)	Wild Birds	Lake
2009.5	H5	2.3.2	Qinghai/Hainan	121(D)	Wild Birds	Lake
2009.4	H5	7.2	Tibet/Lasa	1679(culled)	Chicken	Live bird market
2009.2	H5	2.3.4	Xinjiang/Hetian	519(D)	Goose	Backyard
2008.6	H5	2.3.4	Guangdong/Jiangmen	3873(D)	Duck	Backyard
2008.4	H5	2.3.4	Tibet/Basu	268(D)	Chicken	Backyard
2008.3	H5	2.3.4	Guangdong/Guangzhou	114(D)	Chicken	Live bird market
2008.2	H5	2.3.4	Guizhou/Zunyi	3993(D)	Chicken	Backyard
2008.2	H5	2.3.4	Tibet/Lasa	132(D)	Chicken	Backyard
2008.1	H5	2.3.4	Tibet/Gongga	1000(D)	Chicken	Backyard
2007.12	H5	2.3.4	Xinjiang/Tulufan	4850(D)	Chicken	Backyard
2007.9	H5	2.3.4	Guangdong/Guangzhou	9830(D)	Duck	Backyard
2007.5	H5	2.3.4	Hunan/Yiyang	11172(D)	Duck	Backyard
2007.3	H5	2.3.4	Tibet/Lasa	680/7670	Chicken	Live bird market

# Geographical distribution of outbreaks(2007-2012)



## **Report-2: Active AI surveillance/monitoring program of domestic birds**

- ◆ **Implementation scheme:** The local labs in each province are in charge of the surveillance of avian influenza in poultry farms, any HPAI suspicious samples are subject to be confirmed by the reference lab; the reference laboratory is responsible for the HPAI diagnosis and the active surveillance in live bird markets.
- ◆ **Objectives:** To detect the vaccination coverage rate in the poultry; To discovery and understand the evolution of the AIV and offer the technical basis for controlling the AI.
- ◆ **Target AIV subtype:** H5,H7,H9.
- ◆ **Diagnostic Methods:** Detection of antibody, Virus isolation, Virulence test, and Genetic analysis.
- ◆ **Sampling methods:** Chicken/duck/goose; Blood, tracheal/cloacal swab; Live bird markets, farms, poultry slaughter houses.

# Poultry samples collected from 2009 to 2011

Year	No. Sample (Blood/Tracheal and cloacal swab)	Sampling place	Target premises surveyed
2009	11521/11607	18 Provinces	11 Farms, 10 Poultry slaughter houses, 137 Live bird markets, 1088 Flocks totally
2010	10525/10345	19 Provinces	20 Farms, 7 Poultry slaughter houses, 102 Live bird markets, 975 Flocks totally
2011	11491/10989 (1603/1050)	19 Provinces	26 Farms, 11 Poultry slaughter houses, 106 Live bird markets, 1011 Flocks totally

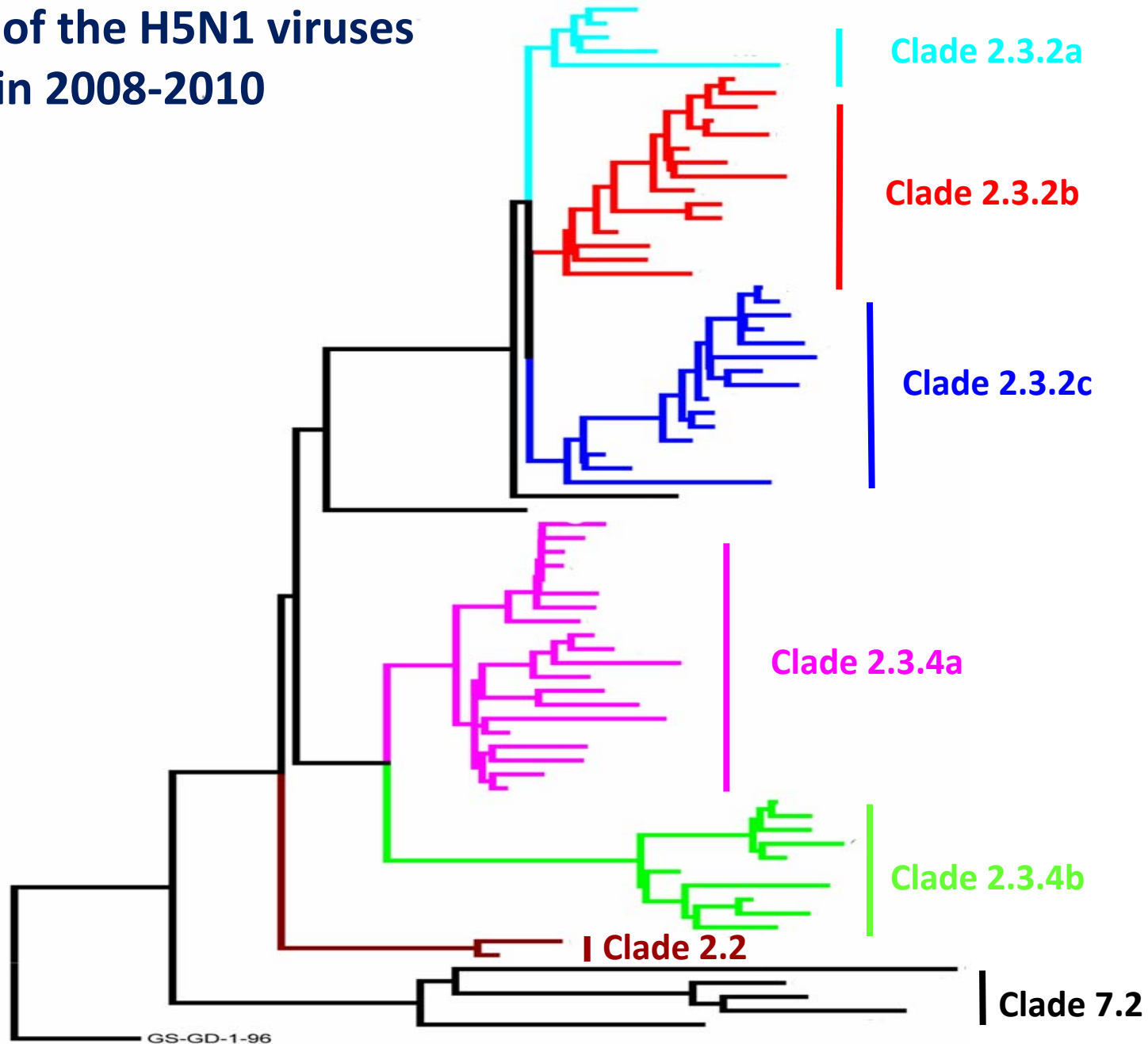
Note: Environment samples/Water samples

## AIVs isolated from 2009 to 2011\*

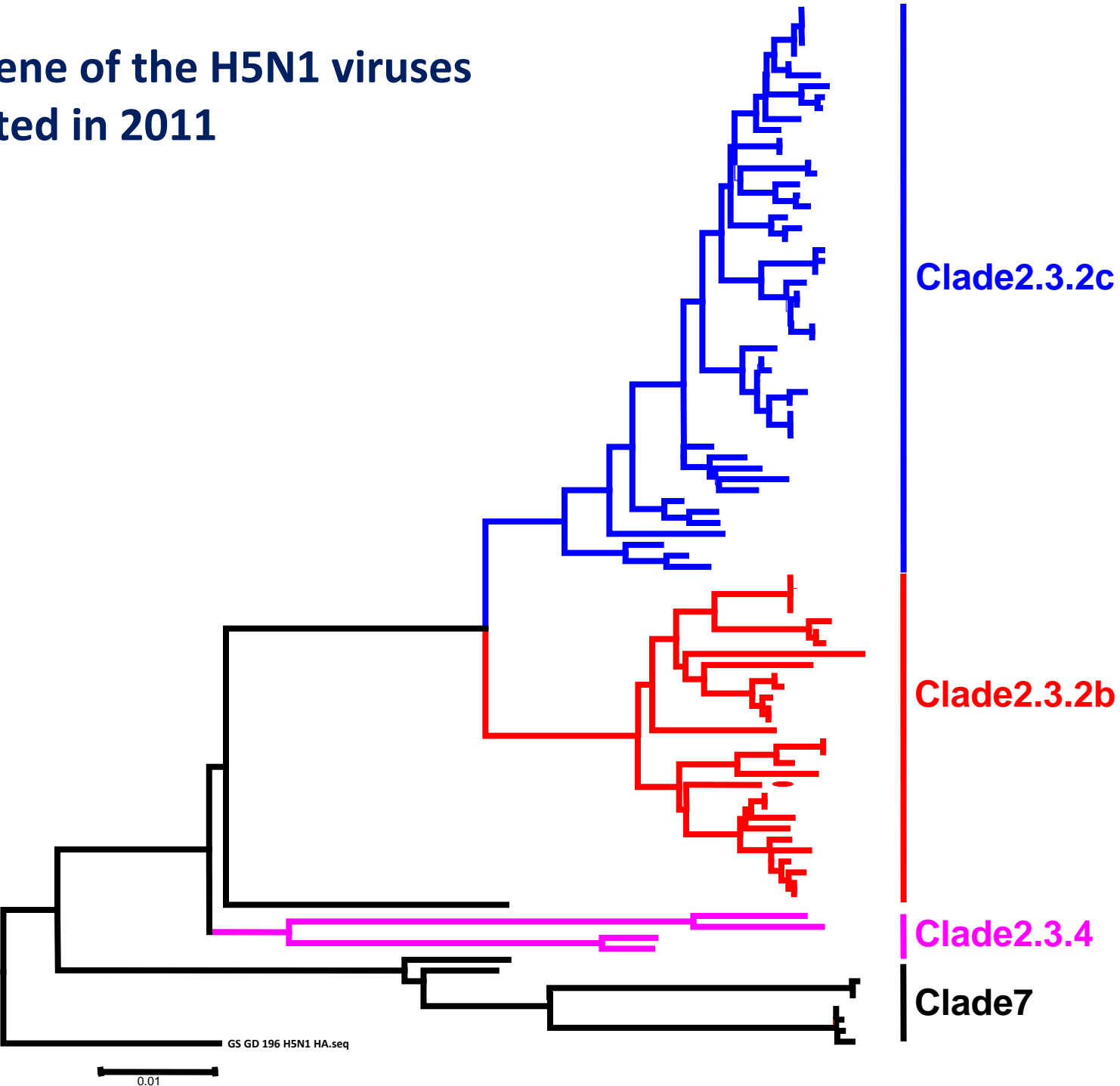
Year	Number of swab samples	Total number of isolates	H5 isolates
2009	<b>11607</b>	667	<b>118</b>
2010	<b>10345</b>	560	<b>87</b>
2011	<b>10989</b>	382	<b>71</b>

\*All H5 viruses were isolated from live bird markets. Samples from poultry slaughter houses and farms are H5 virus free.

# HA gene of the H5N1 viruses isolated in 2008-2010

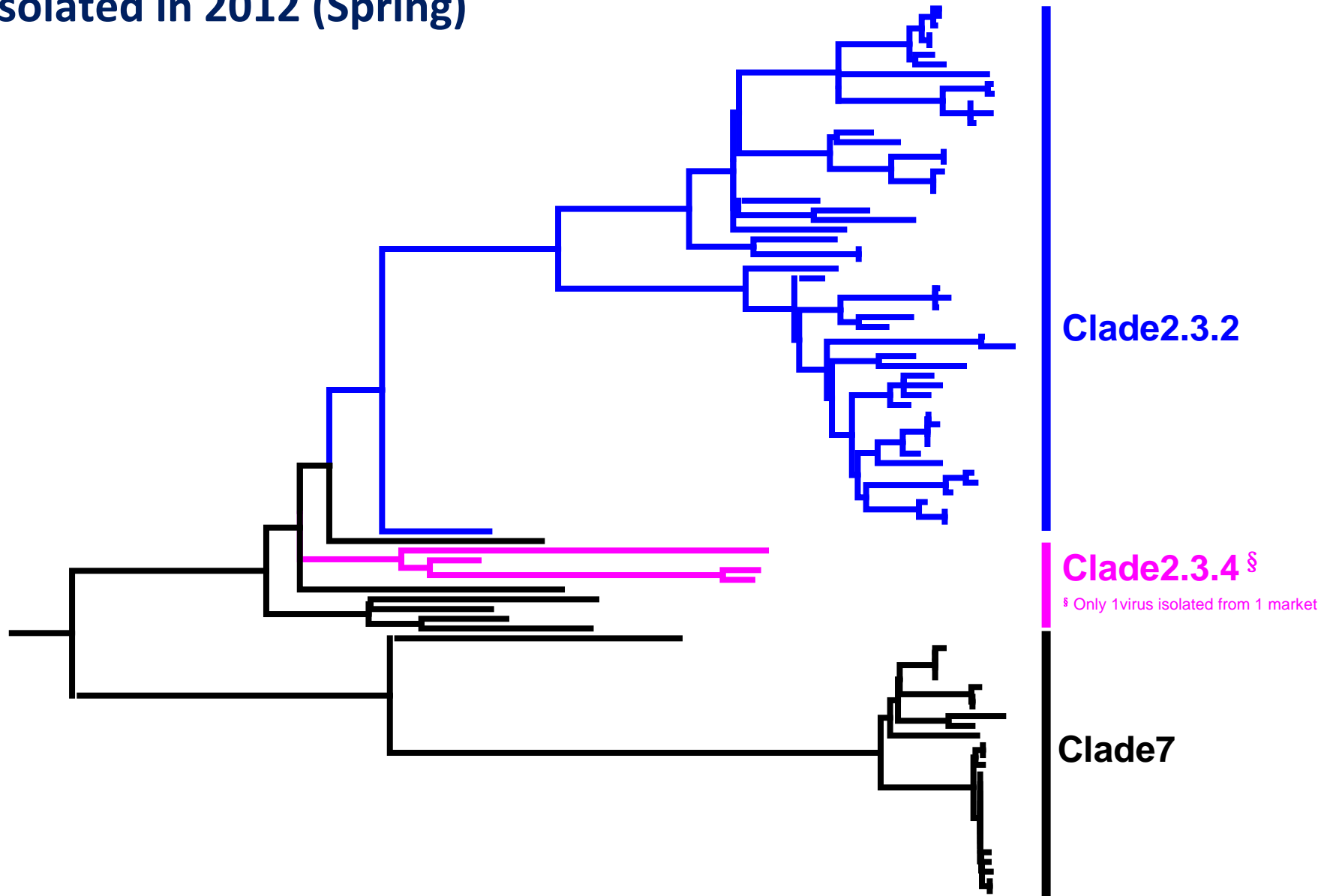


HA gene of the H5N1 viruses  
isolated in 2011





# HA gene of the H5N1 viruses isolated in 2012 (Spring)



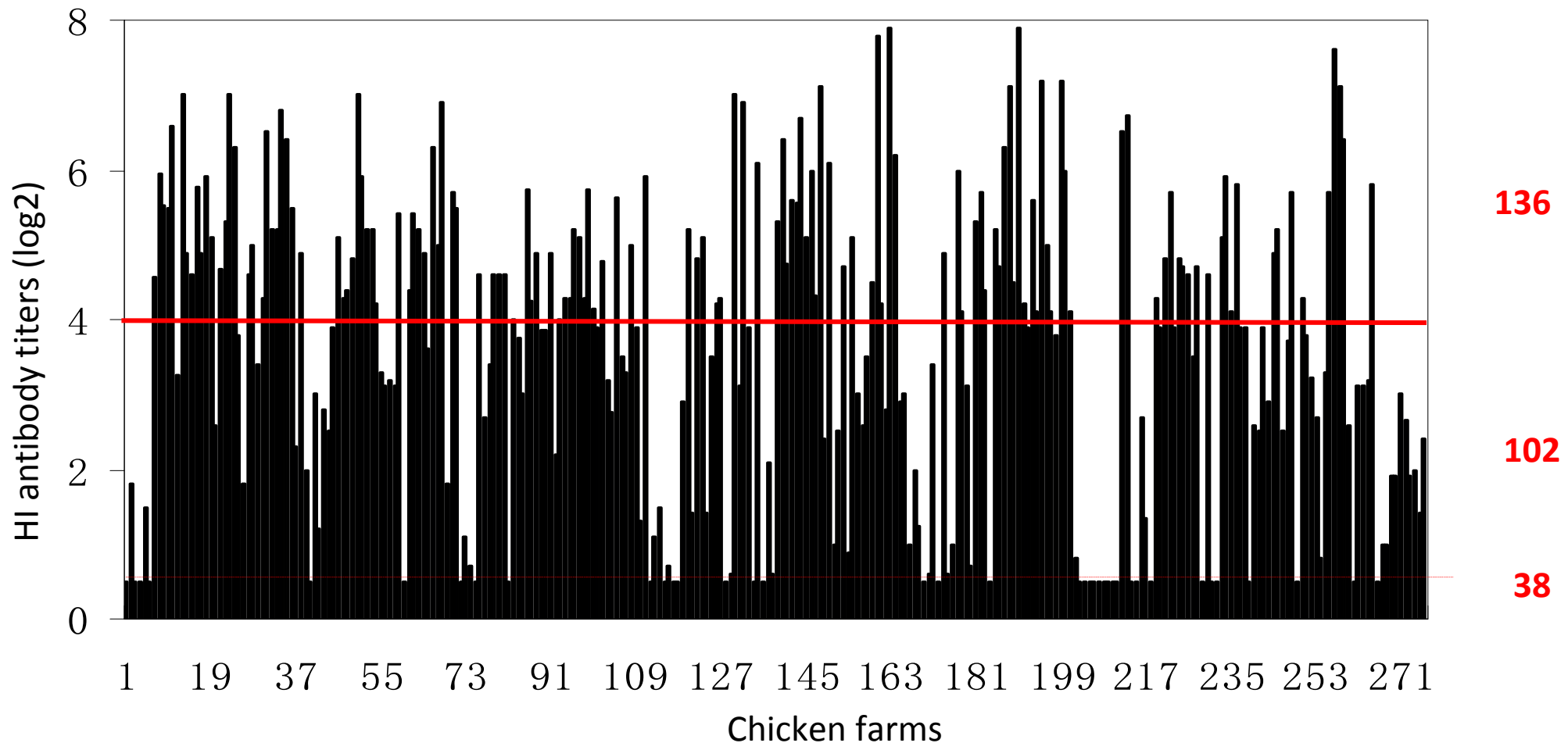
## 2010 spring survey

Time of sampling	No.Sample ( Blood/Tracheal and cloacal swab )	Sampling place	Target premises surveyed
2010. 3. 1-10	4144/4110	12 provinces	45 Live bird markets, 411 flocks

- **45** live bird markets in **12** provinces
- **4110** swabs and **4144** sera samples
- Birds came from **276** chicken farms, **114** duck farms and **18** goose farms (**408**)

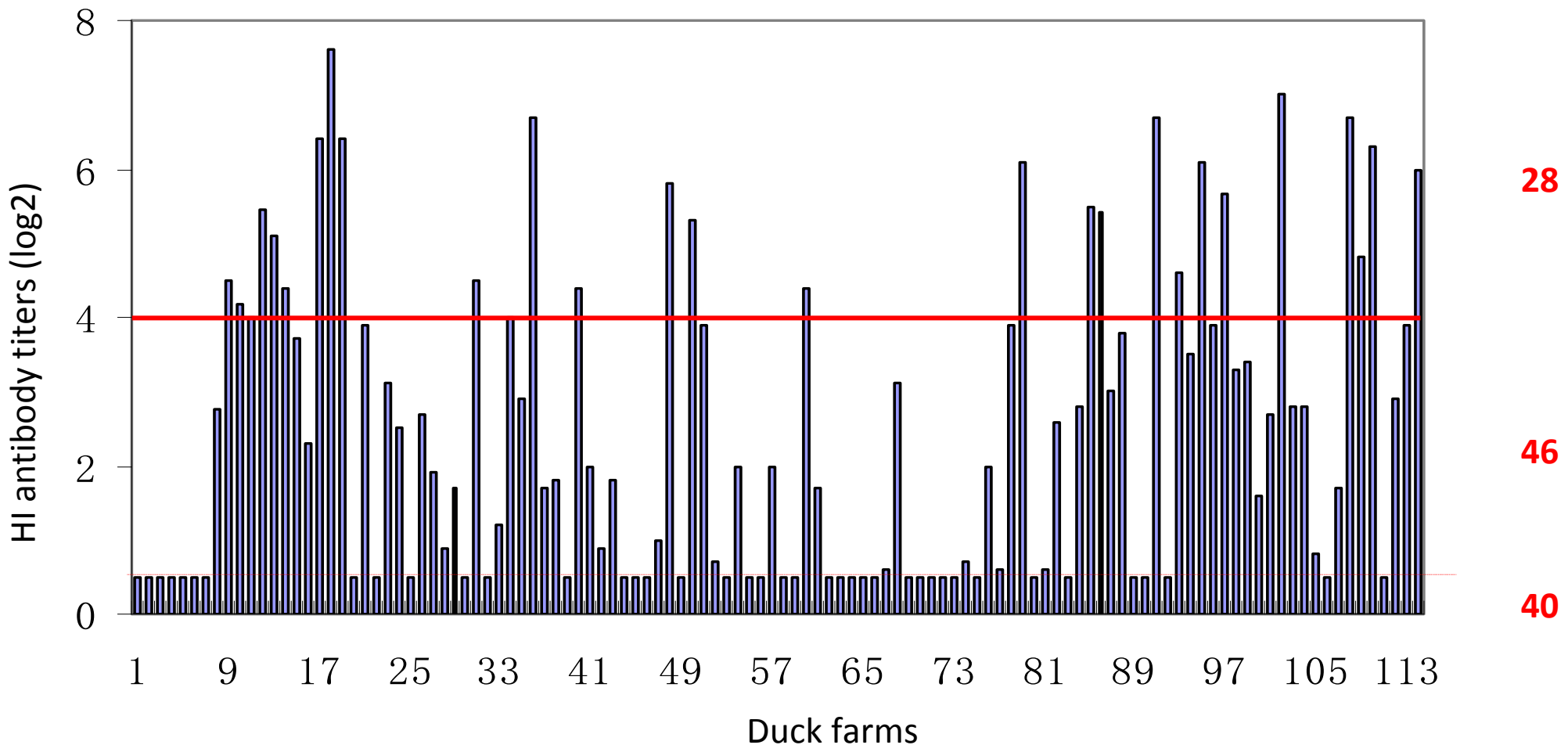
# H5 antibodies of **chicken flocks** in 2010 spring survey

**136** out of **276** (49.2%) chicken farms were qualified vaccinated, while  
**38** out of **276** (13.7%) farms were not vaccinated



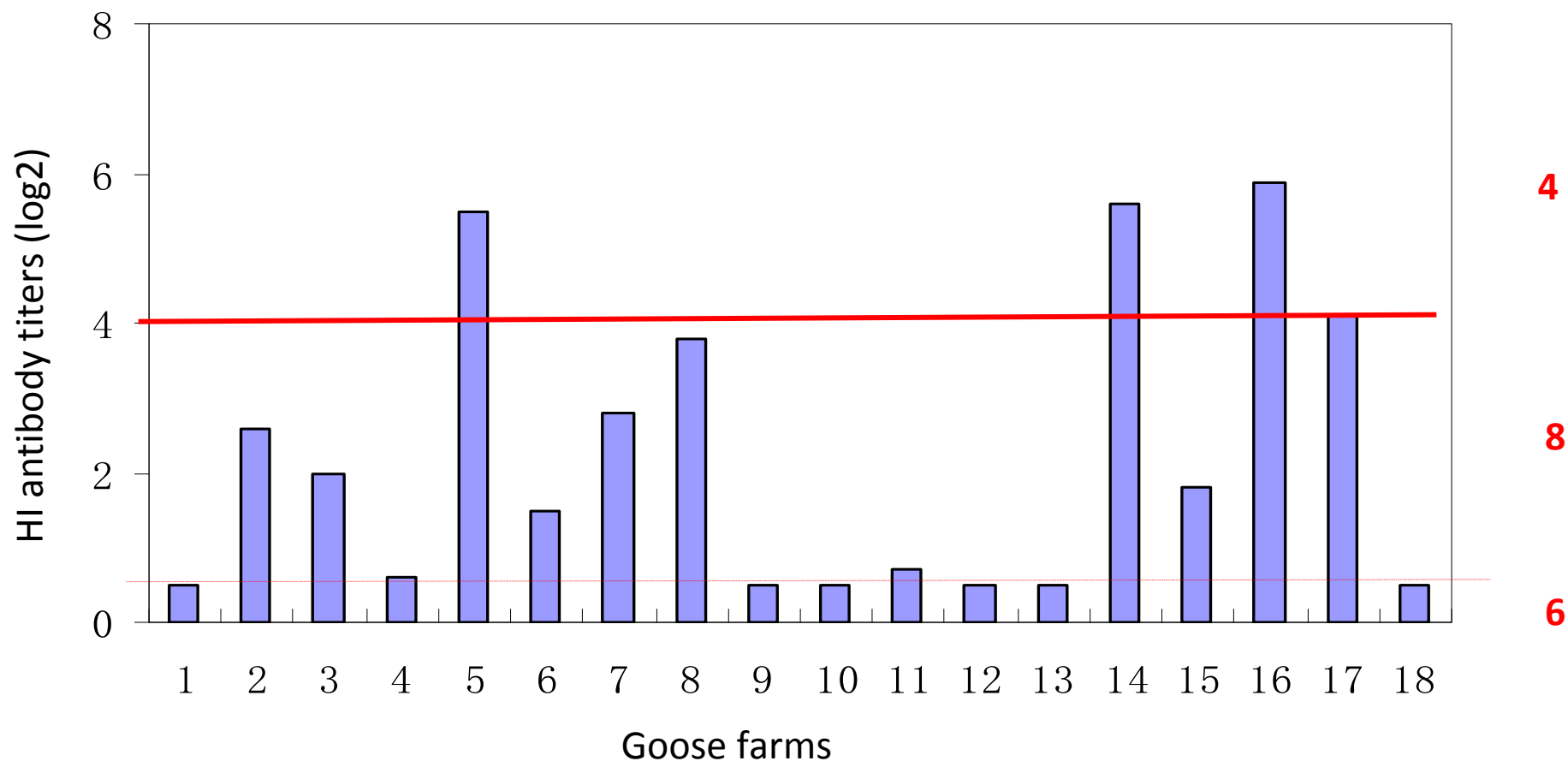
# H5 antibodies of **duck flocks** in 2010 spring survey

**Only 28** out of **114** (24.5%) duck farms were qualified vaccinated, while **40** out of **114** (35%) farms were not vaccinated



# H5 antibodies of **goose flocks** in 2010 spring survey

**4** out of **18** goose farms were qualified vaccinated, while **6** of them were not vaccinated.



## Serological analysis of 2010 spring survey

- ❑ 136 out of 276 (49.2%) chicken farms were qualified vaccinated, while 38 out of 276 (13.7%) farms were not vaccinated
- ❑ Only 24.5% duck farms were qualified vaccinated, while 35% farms were not vaccinated
- ❑ 4 out of 18 goose farms were vaccinated, while 6 of them were not vaccinated
- ❑ All of the birds that have qualified H5 HI antibody titers were H5 influenza virus negative

# Summary

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- Clade 2.3.2 viruses are in a dominant position in China and clade 7 viruses still could be detected in the chickens in some areas in northern China
- The vaccination is playing important roles to protect poultry from H5 influenza infection
- Vaccine coverage rate needs to be increased to completely control or eradicate the disease, though improved biosecurity, extensive surveillance and an effective monitoring program are always important.

# Report-3: Active/passive AI surveillance/monitoring/investigation programme of wild birds

## New H5N1 AIVs in wild birds, Qinghai, China, 2009

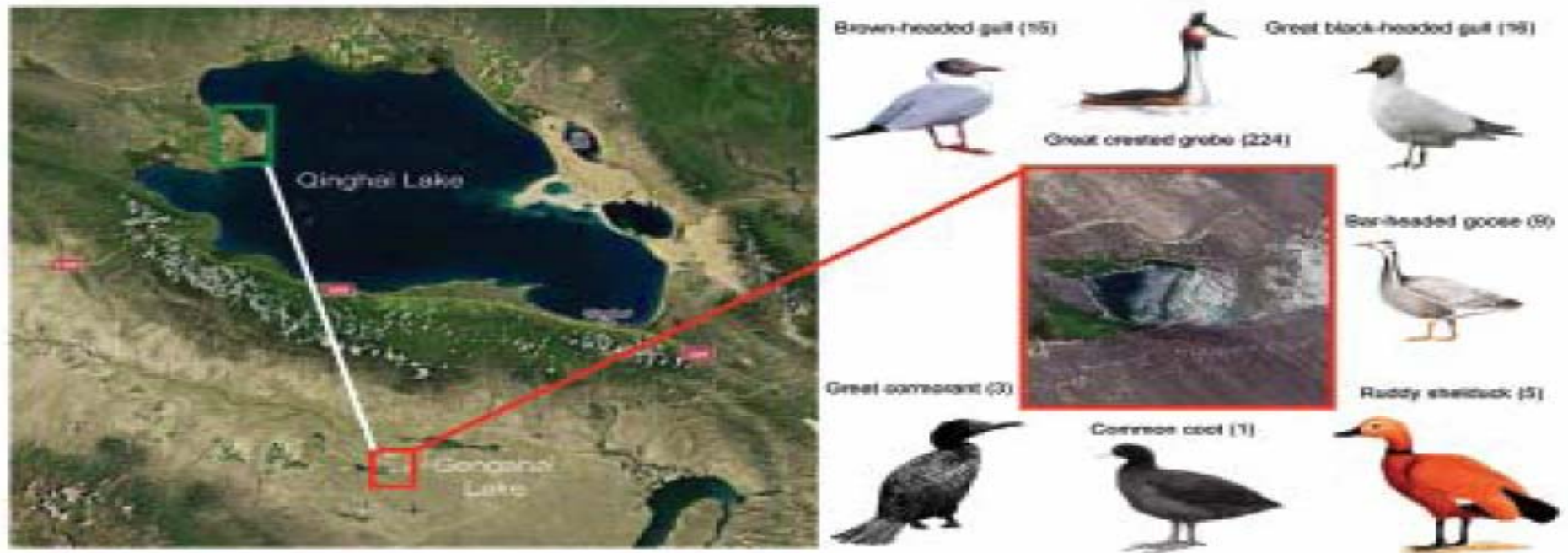


Figure 1. Location in Qinghai, China, of dead birds that were tested for avian influenza virus (H5N1), with images and common names of bird species tested. Red box indicates Gengahai Lake, where dead birds were detected, and green box indicates Bird Islet of Qinghai Lake; the distance between them is 90 km. Numbers of dead birds of each species are indicated in parentheses.



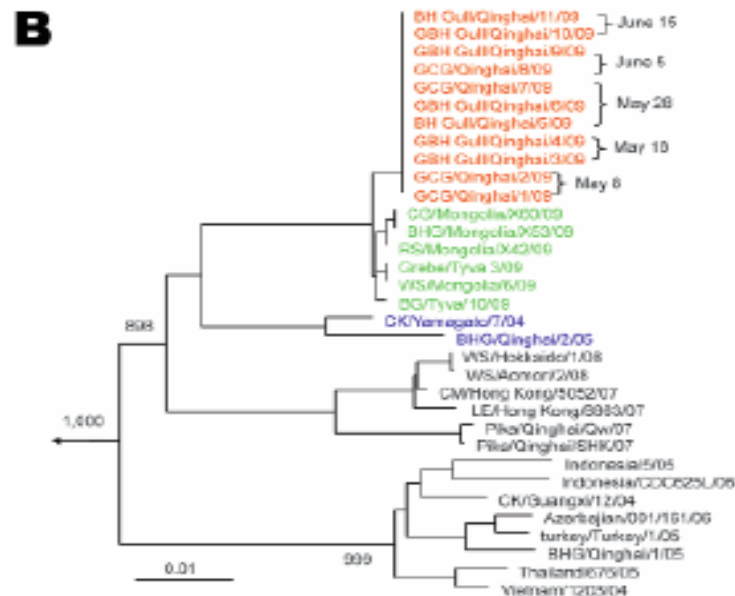
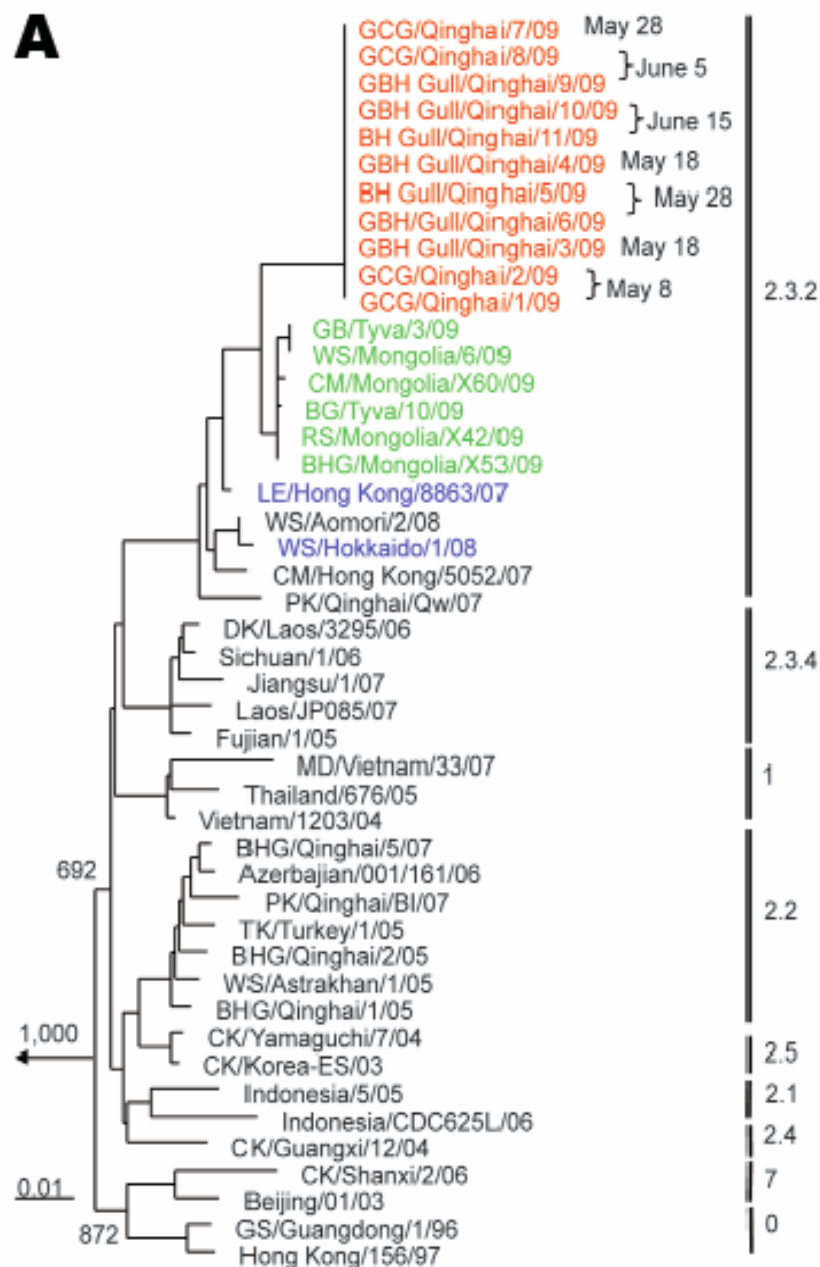


Figure 2. Phylogenetic trees of hemagglutinin genes (nt 29–1,728) (A) and acidic polymerase genes (nt 25–2,151) of avian influenza viruses (H5N1) (B). Clade numbers are indicated on the right in panel A. Trees were constructed by using the PHYLIP program of ClustalX software version 1.81 ([www.clustal.org](http://www.clustal.org)), the neighbor-joining algorithm, and rooted to A/chicken/Pennsylvania/1/83(H5N2). Bootstrap analysis was performed with 1,000 replications. Viruses obtained in this study are shown in red, previously detected viruses that are closely related to avian influenza virus (H5N1) QH09 are shown in blue, and closely related viruses that were detected after the Qinghai wild bird outbreak in 2009 are shown in green. Dates of virus isolation are shown. Scale bars indicate nucleotide substitutions per site. GCG, great crested grebe; GBH, great black-headed gull; BH, brown-headed gull; GB, grebe; WS, whooper swan; CM, common magpie; BG, bean goose; RS, ruddy shelduck; BHG, bar-headed goose; LE, little egret; PK, pike; DK, duck; MD, Muscovy duck; TK, turkey; CK, chicken; GS, goose; CG, common goldendye.

## 2013 forward

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- ❑ Active surveillance of wild birds is hard to imply
- ❑ Effective strategy must be applied to strengthen the wild bird surveillance along Migratory Flyways
- ❑ Promote the collaboration with State Forestry Administration for preventing the transmission from wild birds to domestic birds



**Department of Animal Health and Production  
Ministry of Agriculture, Forestry and Fisheries**



## **Controlling and Prevention Measures of H5N1 HPAI in domestic birds and wild birds in Cambodia**

Tokyo, Japan 13-14 December 2012

# Avian Influenza

- 80% of Cambodians live in rural area
- Raising poultry for livelihood
- Poultry and Humans
- H5N1 outbreaks in many Asian countries since late 2003
- 33 poultry outbreaks and 21 human cases in Cambodia to date

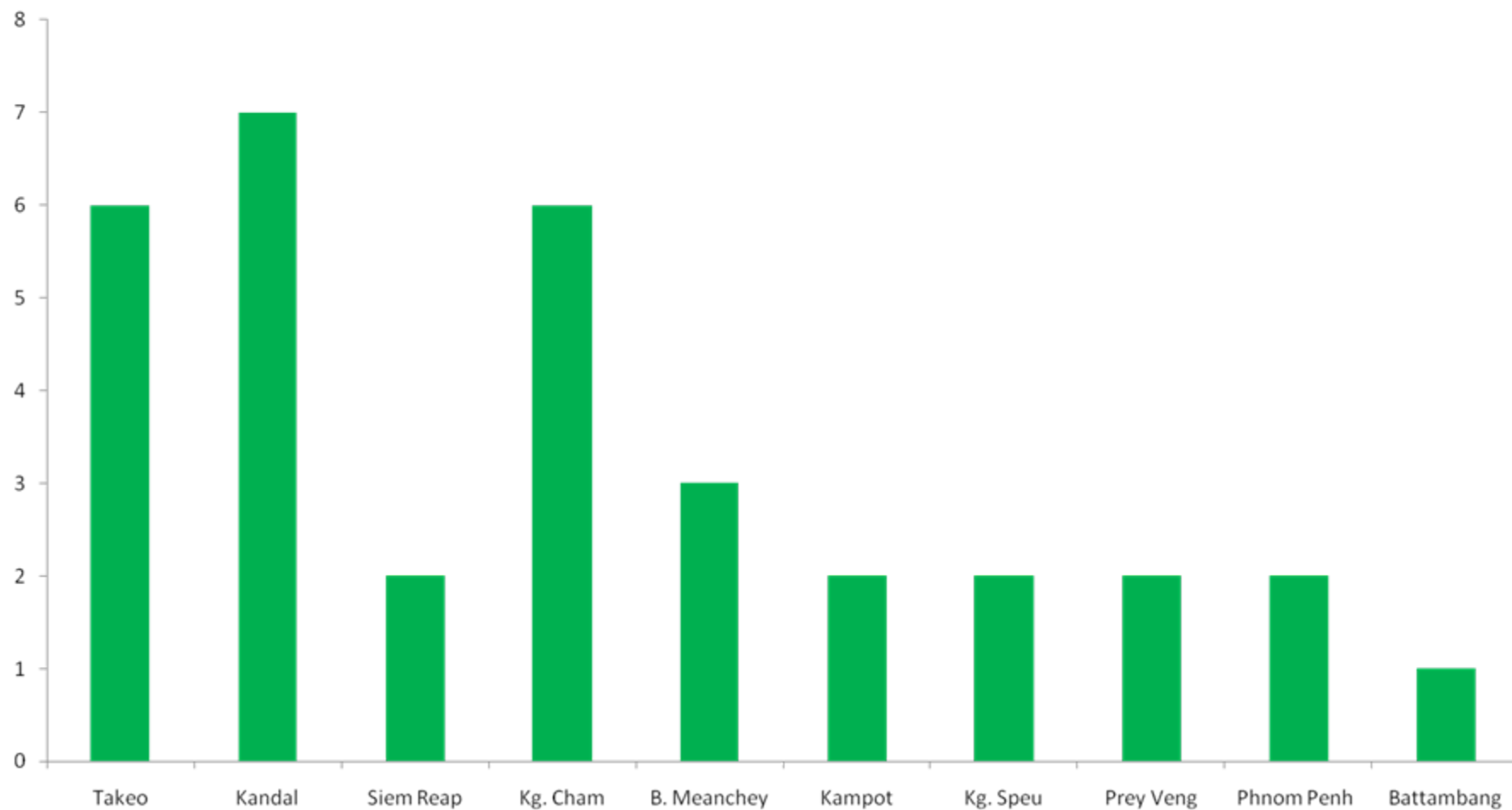
# 33 H5N1 Outbreaks

- Takeo: 06
- Kandal: 07
- Siem Reap: 02
- Kampong Cham: 06
- Banteay Meanchey: 03
- Kampot: 02
- Kampong Speu: 02
- Prey Veng: 02
- Phnom Penh: 02
- Battambang: 01

**This year 2012, we had 01 H5N1 outbreaks.**

**The last outbreak on May 31, 2012 in Kampong Speu province**

## Number of H5N1 outbreaks by province (2004-2012)



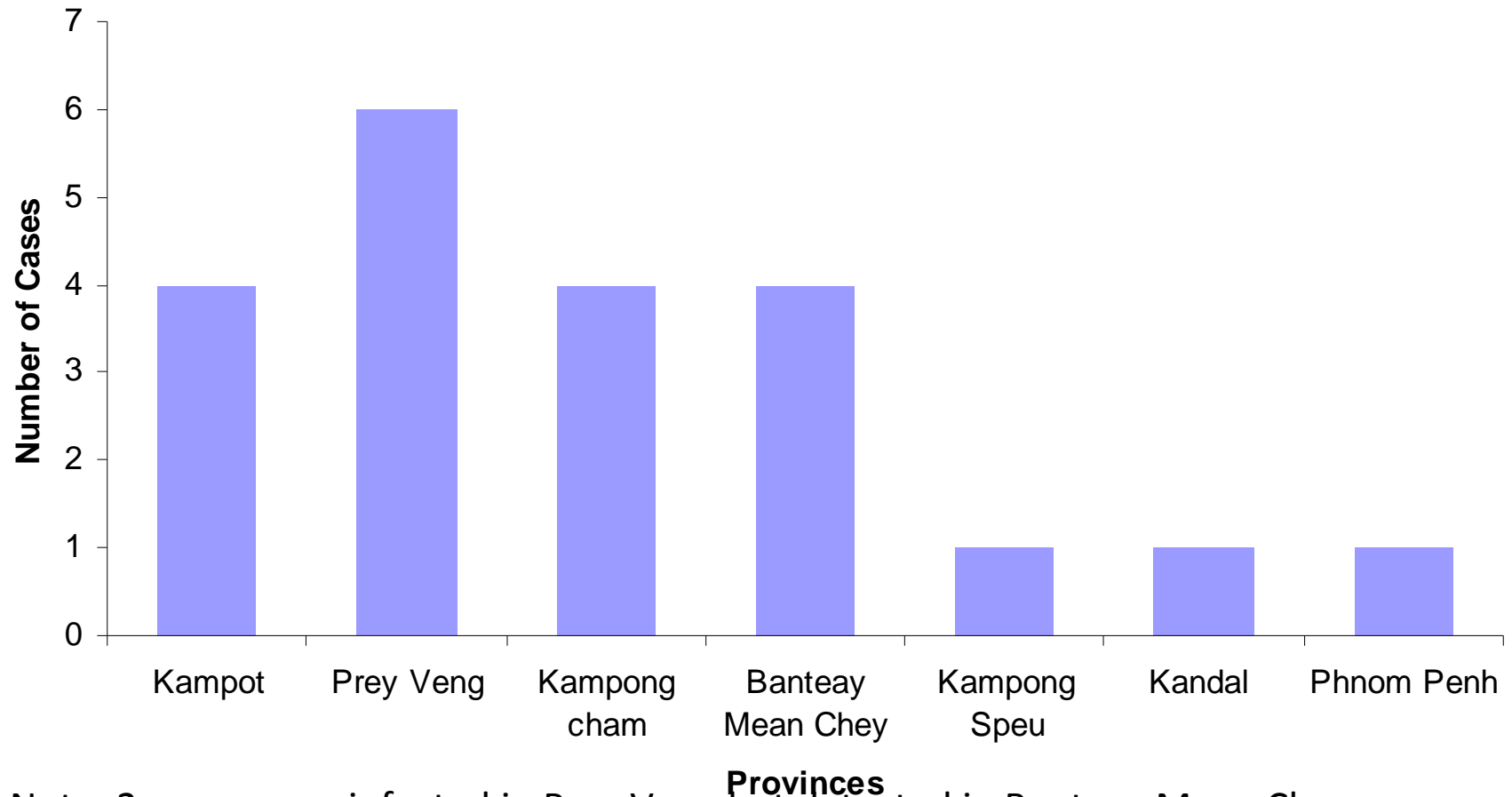
# Human cases of H5N1

- 2005: 4 cases (4 deaths) in Kampot
- 2006: 2 cases (2 deaths) in Kampong Speu and Prey Veng
- 2007: 1 case (1 death) in Kampong Cham
- 2008: 1 case (alive) in Kandal
- 2009: 1 case (alive) in Kampong Cham
- 2010: 1 case (death) in Prey Veng
- 2011: 8 cases (death)
- 2012: 3 cases (death)

**Total: 21 human cases (2 alive)**

# Human Avian Flu A/H5N1 from 2005-Present (N=19)

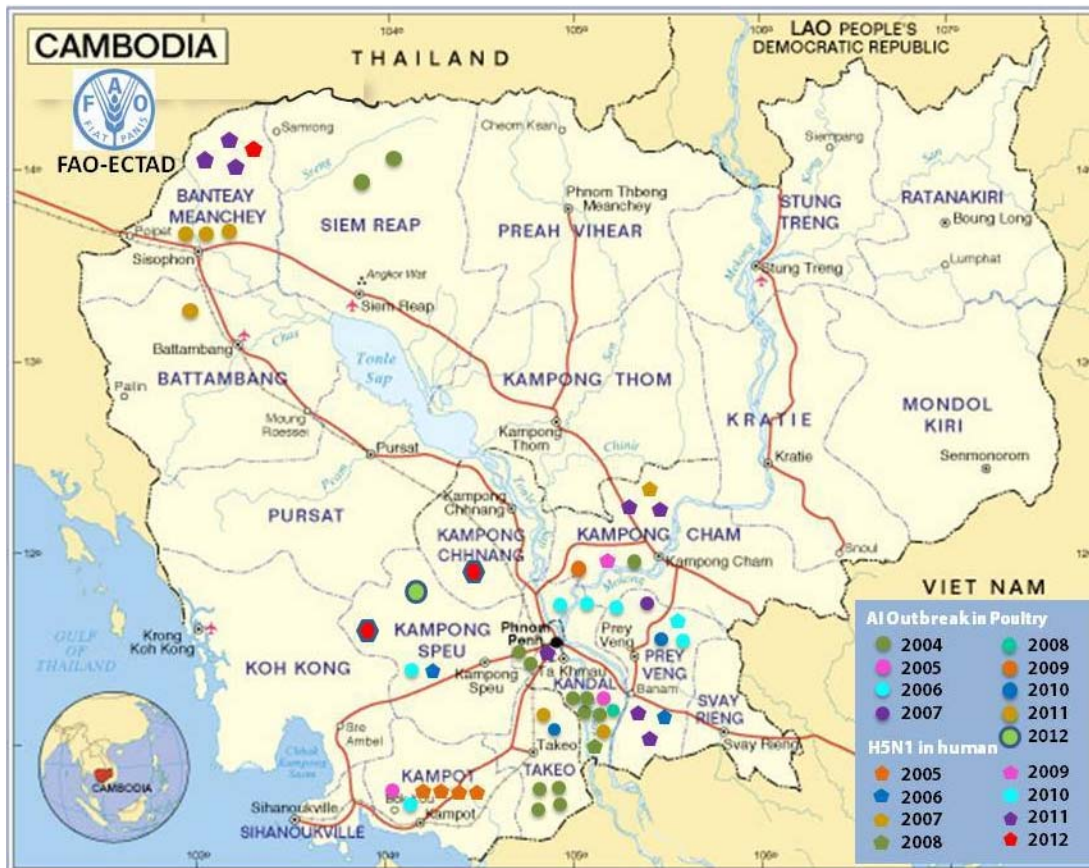
Human Avian Flu A/H5N1 Cases by Province



Note: 2 cases were infected in Prey Veng but detected in Banteay Mean Chey



# Distribution of HPAI (Human and Poultry) Outbreaks, January 2004 - August 2012



33 poultry outbreaks / 21 human cases (19 fatal)

# Laboratory Confirmation of H5N1

- Serology

- HA and HI Test

- Anti-influenza A (ARGENE, France)

- H5N1 Antigen (UK)

*or*

- Virus Isolation

- Egg Inoculation Test

- H5N1 Antiserum (UK)

- Real Time PCR

# Virus characteristics

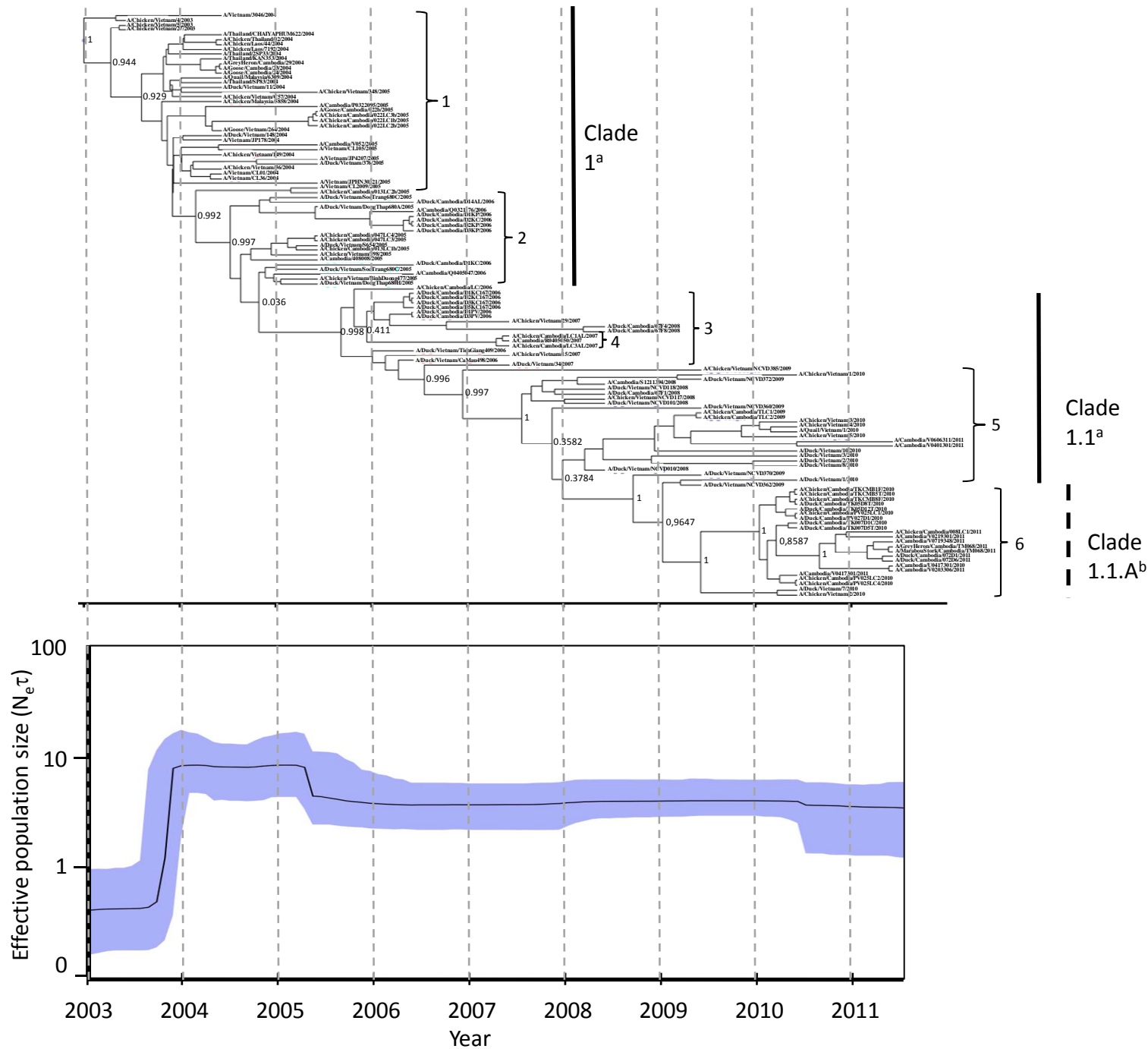
- All viruses isolated from human, poultry and wild bird since 2004 until now is belonged to **clade 1 and clade 1.1**
- Analyses do not show evidences of better adaptation to human, nor resistance to oseltamivir but all Cambodian viruses are **resistant to amantadine/rimantadine**. Some mutation associated with **high virulence** for human and other mammals were detected in some Cambodian strains
- In 2007, isolation of a strain that showed an antigenic drift → this virus is include in the pre-pandemic human **H5N1 vaccine**

# Virus characterists

- 6 lineages belonging to clades 1 and 1.1 were identified since 2004
- Lineage 1 shares aj ancestor with viruses from Thailand and disappeared after 2005
- Lineage 2 replaced lineage 1 and originated from Vietnam
- Lineage 3 replaced lineage 2
- Lineage 4 was seen only in Cambodia (2007; strain included as potential seed virus in pre-pandemic vaccine)

# Virus characteristics

- Lineage 5 is circulating both in Cambodia and Vietnam since 2008
- Lineage 6 seems to be endemic to Cambodia since 2010 and was suggested to be named clade 1.1.A
- Lineages 5 and 6 were co-circulating in 2011 indicating at the same time transmission specific to Cambodia and circulation of strains probably introduced through illegal poultry trade



**Figure 2**






# AI surveillance on domestic birds and wild birds

**Active surveillance** is based on specific targeted investigation of at-risk populations for evidence of infection that may be based on detecting exposure to the agent (antibody detection by serology) or the presence of the agent (virus or antigen detection).

**Passive surveillance** is based on notification of suspected disease; it depends on villagers, farmers, private vets, VC, VAHWs to report disease to veterinary services

# Active surveillance on domestic birds

5 main activities are implemented in Cambodia

-  Sentinel surveillance in free-grazing duck farms/flocks
-  Market surveillance on live ducks
-  Environmental sampling in markets
-  Surveillance when outbreaks occur
-  Sample testing



# Sentinel surveillance in free-grazing duck farms/flocks

## Activities implemented

- Firstly, surveillance has been organized from March to October 2010
- 12 duck commercial farms/flocks in Takeo, Kampong Cham, Prey Veng, Kampot, Battambang and Preah Sihanouk provinces
- 400 ducks tagged with a ring
- Ducks are collected every two weeks

# Market surveillance on live ducks

- Surveillance in the markets started in 2006 organized in Phnom Penh (Demkor and Orussei), Siem Reap, Svay Rieng, Battambang, PreahSihanouk, Pursat, B. Meanchey, Kampong Cham , Kandal, Takeo, Kampot.
- NaVRI teams assisted by provincial staffs collect tracheal and cloacal swab and blood samples
- Market are sampled twice a month



# Environmental sampling

- 502 samples have been collected in 4 markets in Kampong Cham, in Phnom Penh and in Takeo.
- 147 were water samples, 120 feathers, 117 feces and 118 mud and soil samples
- **37% of water samples, 25% of feathers, 14.5% of feces, contain H5N1 virus (RT-PCR method)**

# Surveillance when outbreaks occur

- When outbreak occur in poultry flocks or when human cases are detected, the NaVRI staff investigate and conduct active surveillance in the area.
- Firstly, the teams make a complete survey in the village where the H5N1 outbreak is identified and collect samples on any sick poultry, on poultry in the neighboring houses.
- The second step is to visit neighboring villages, especially if some mortality/morbidity is reported; NaVRI teams collect samples on diseased animals

# Surveillance when outbreaks occur

## Example

- On 04 February, the 12<sup>th</sup> human case has been confirmed on a 5-year boy leaving near Phnom Penh in Prek Leap.
- NaVRI conducted investigation and active surveillance resulting in 90 samples collected in the village. All samples have been found negative

# Active surveillance on domestic birds

- Only one NGO named Wildlife Conservation society (WCS) cooperated with NaVRI to conduct surveillance on HPAI in wildbird (Sparrow species)
  - Results shown that all samples were negative of H5N1 virus





**Thank you for you attention !**



## IMPLEMENTATION OF THE PROGRAMME ON SURVEILLANCE OF WILD AND DOMESTIC BIRDS ALONG MIGRATORY FLYWAYS



**BHUTAN**

*Dr. N. Dahal*

*Deputy CVO*

The 5th OIE Regional Expert Group Meeting for Implementation of the Programme on Surveillance of Wild and Domestic Birds along Migratory Flyways,  
Tokyo, Japan, 13-14 December 2012)





# Report-1:

## Recent HPAI outbreaks and H5N1 HPAI virus characterization studies in domestic birds and wild birds

### 1. Outbreaks & premises affected

Year	Number of outbreaks /locations	Species affected	Premises affected	Remarks
2010	01/ 03	Domestic poultry	Backyard	February 2010
2012	03/ 12 01/01	Domestic poultry, pigeon and sparrow	Backyard /semi-commercial	January 2012 October 2012



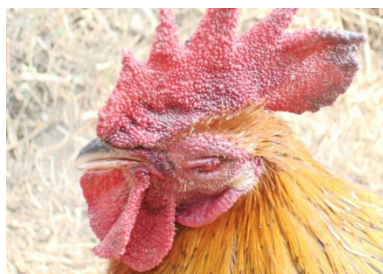
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# Report-1: Continued.....



Year	Poultry destroyed	Villages affected	Households involved	Amount compensated (poultry, eggs, coops & feed)
2010	5379	25	317	6,97,516.00 (USD~ 12916)
2012 (January)	3086	19	215	905335.00 (USD ~16765)
2012 (October)	31	4	8	3139.00 (USD~ 59)



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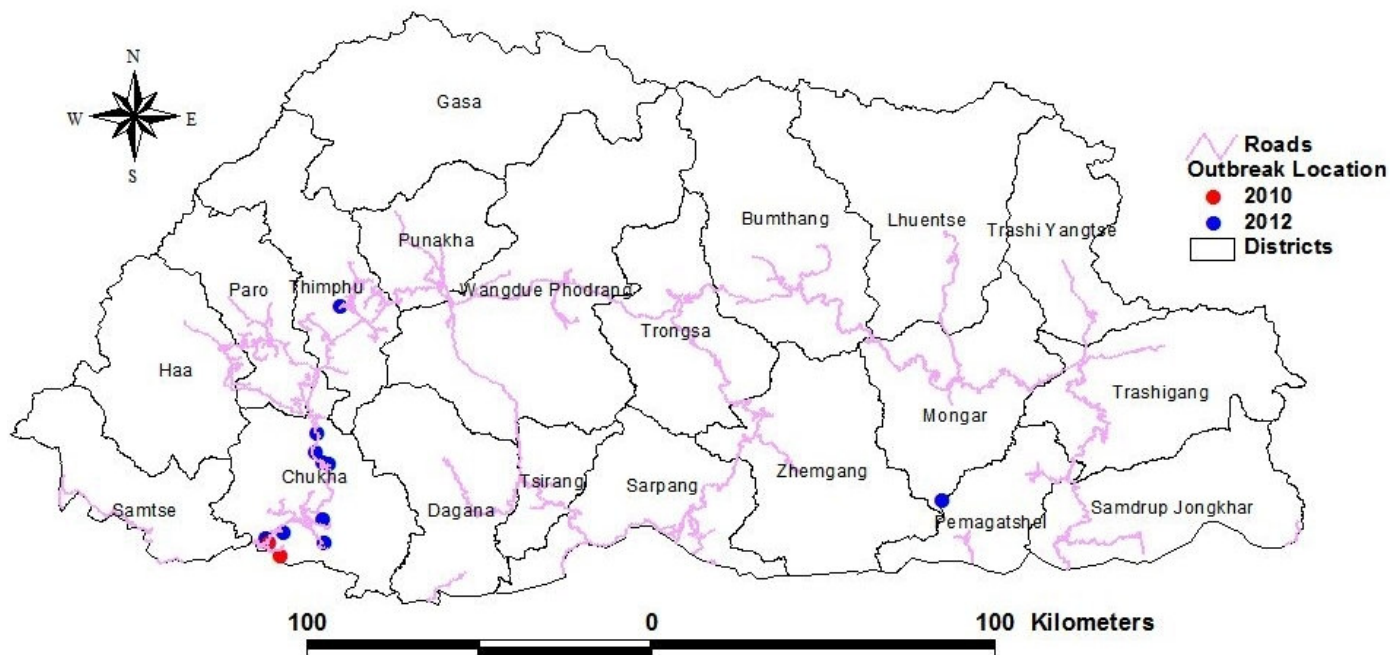




# Report-1: Continued.....



## 2. Geographical distribution of outbreaks



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# Report-1: Continued.....



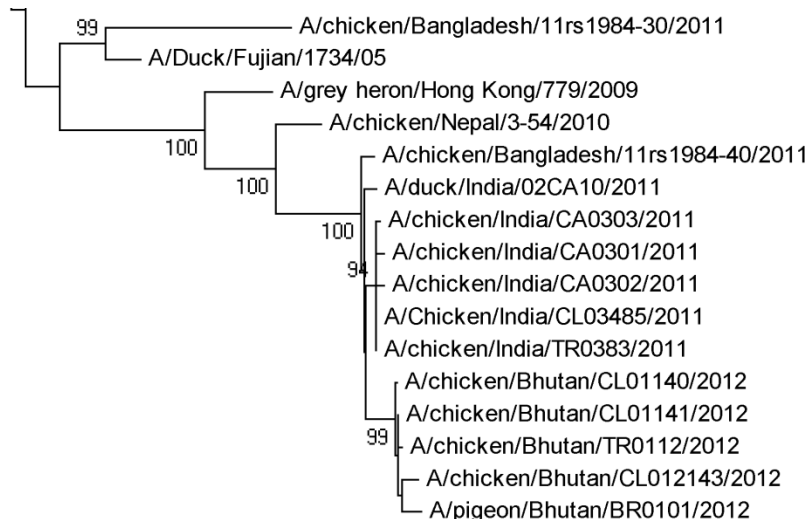
## 3. Information on the virus isolates

**Subtype: A/H5N1**

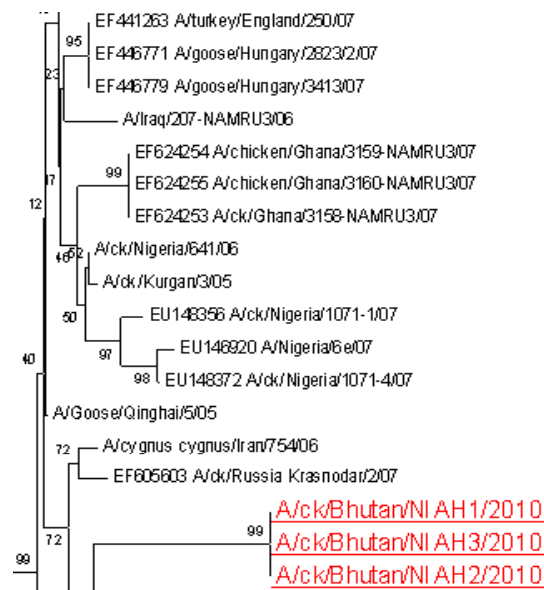
**2010 –Virus clade 2.2**

**2012 – Virus clade 2.3.2.1**

- Similar to isolates from Bangladesh & India
- Sensitive to Oseltamivir & Amantadine



2.3.2.1



Clade 2.2



## Report-1: Continued.....



### Information on 2010 A/H5N1 Bhutan isolate

- Isolate – A/H5N1 “Qinghai-like” belonging to clade 2.2 (subclade 2.2.3)
- Isolate highly virulent to chicken (IVP Index – 2.88)
- Complete genomic analysis indicated 99.1-100% homology with isolates from Bangladesh
- None of the isolates had mutations in the gene coding for the M2 ion channel or NA protein that confer resistance to amantadine and oseltamivir.
- All the Bhutan isolates possessed amino acid K at position 627 of PB2 protein, which is associated with increased virulence in mice and adaptation to humans
- 2010 Bangladesh isolate shared the unique molecular markers with Bhutan isolates and also grouped with them with 100% bootstrap value.
- Bhutan isolates contain additional molecular marker at <sup>678</sup>N in PB1 as compared to Indian and other Bangladesh isolates.

**Ref:** Dubey et al, (2011): Isolation and characterization of influenza A virus (subtype H5N1) that caused the first highly pathogenic avian influenza outbreak in chicken in Bhutan. *Veterinary Microbiology*, 155(2012) 100-105



## Report-2:

# Active AI surveillance/monitoring programme of domestic birds (Serological and virological surveillance)

## 1. Active surveillance in domestic poultry/waterfowls

- HPAI is a notifiable disease in Bhutan (Livestock Rules & Regulations 2008).
- The National Influenza Pandemic Preparedness Plan (NIPPP) mandates active surveillance by the concerned agencies.
- The main objectives of active surveillance (clinical, laboratory & targeted)
  - Enable early detection and rapid response (early warning).
  - Understand the epidemiology of AI and its socioeconomic impact.
  - To help design effective HPAI control programs.
  - To ensure freedom from clinical disease and absence of infection in the country.
  - To evaluate the existing disease control programs.



## Report-2: Continued.....



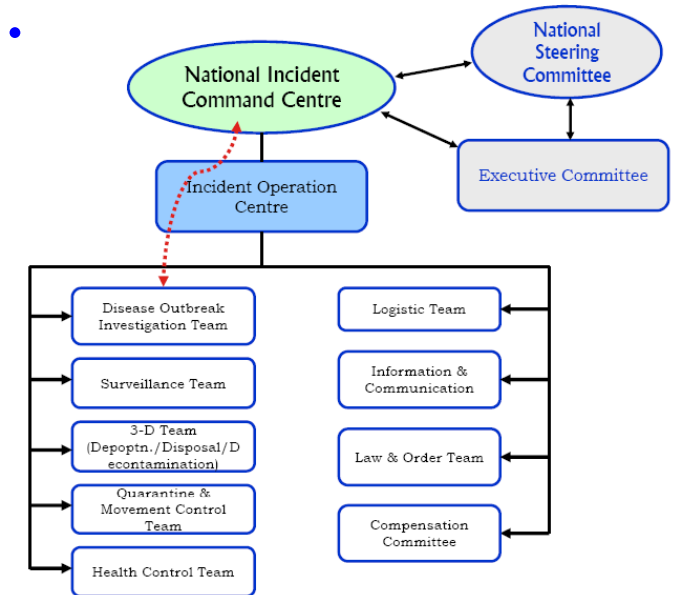
### 2. Surveillance teams

- Veterinary Vigilance team**

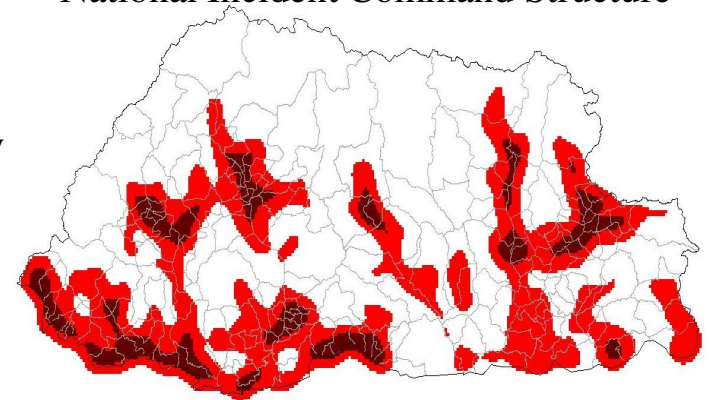
- Regional teams under the Department of Livestock, led by veterinarian and carry out laboratory, clinical & targeted surveillance.
- Advocacy, awareness and sensitization to all relevant stakeholders
- Instant communication to National Centre for Animal Health (NCAH) in any suspicious cases
- Submit weekly report to NCAH

- Border Vigilance team**

- Teams from Bhutan Agriculture & Food Regulatory Authority (BAFRA) stationed at entry points
- Regulate import of livestock products import
- Maintain stringent border vigilance and surveillance



National Incident Command Structure



AI incursion risk score map



## Report-2: Continued.....



### 3. Diagnostic methods

- Target AIV subtype – H5N1 (few samples tested against H7 & H9)
- Field diagnosis
  - Rapid test kits (FluDetect – Synbiotics Corporation, San Diego, CA, USA)
  - H5-HA - Animal Genetics, Inc. Woncheon-dong Korea
  - Type A & H5-HA - Quicking Biotech Co., Ltd., Shanghai, China
- Regional Veterinary Laboratories
  - Real time PCR (eastern region)
- National Veterinary Laboratory
  - HA-HI (H7 & H9)
  - ELISA – IDEXX
  - Real time PCR
- VI & Cell culture facilities in pipeline





## Report-2: Continued.....



### 4. Sampling methods

- Poultry species – primarily chicken & ducks
- Target premises are commercial, semi commercial, backyard and free ranging poultry birds and ducks.
- Targeted sample of around 2500 samples annually (cloacal, tracheal swabs and blood serum)
- Regular sampling on region wise allocation of annual sample targets
- Surveillance is intensified between October – March
- All surveillance samples subjected to rRT-PCR at NCAH
- All positive samples referred to OIE designated laboratory in India (Bhopal) and St Jude CIERS, USA



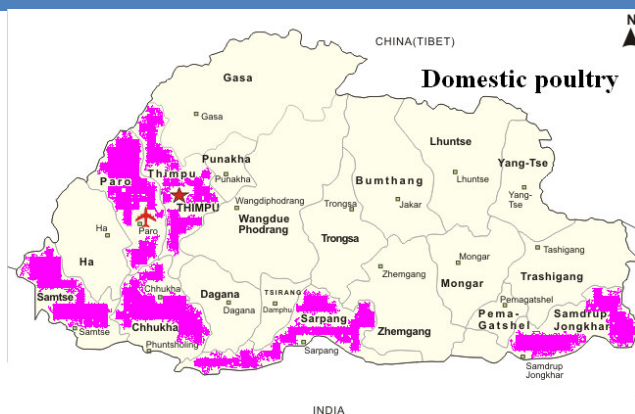
## Report-2: Continued.....

### 5. Results/Findings

Year	Animal spp.	Sample type		No. of samples	Diagnosis		
		*Swabs	Blood serum		Type A	H5-HA	rRT-PCR
2010-2011	Poultry/ Ducks	168	133	301	102	67	171
	Swine	92	56	148	0	0	0
2011-2012	Poultry/ Ducks	327	197	524	221	97	319
	Swine	68	42	110	0	0	0

\*Cloacal/Tracheal/Nasal/

- 11/20 districts sampled
- Total of 1083 samples processed



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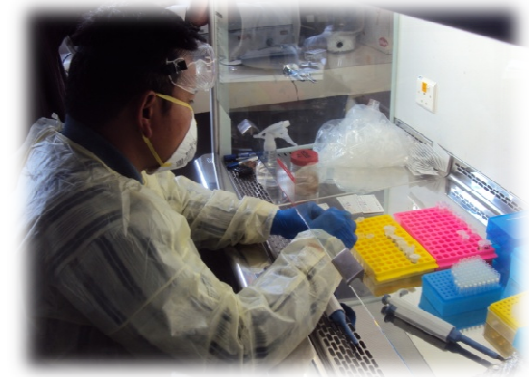


## Report-2: Continued.....



### Outbreak samples

Year	District	*Positives
2010	Chukha	15
2012	Chukha	40
	Thimphu	7
	Mongar	2
*confirmed by OIE referral laboratory		





## Report-3:

# Active/passive AI surveillance/monitoring/investigation of wild birds

## 1. Surveillance in wild birds

- Investigation of unusual mortalities/die-backs in wild birds
- Protocol adapted -FAO manual “**Wild Bird HPAI Surveillance**”
- Bhutan falls under central Asian flyway
- Targeted species of wild birds
  - Black-necked cranes, Ruddy Shell duck, Egrets, Black Storks, Fish eagles, white/grey bellied herons, cormorants, crows and pigeons
- Collaborating partners for surveillance
  - Wildlife Conservation Division of Department of Forests & Park Services
  - Royal Society for Protection of Nature
- St. Jude CEIRS, Memphis, USA





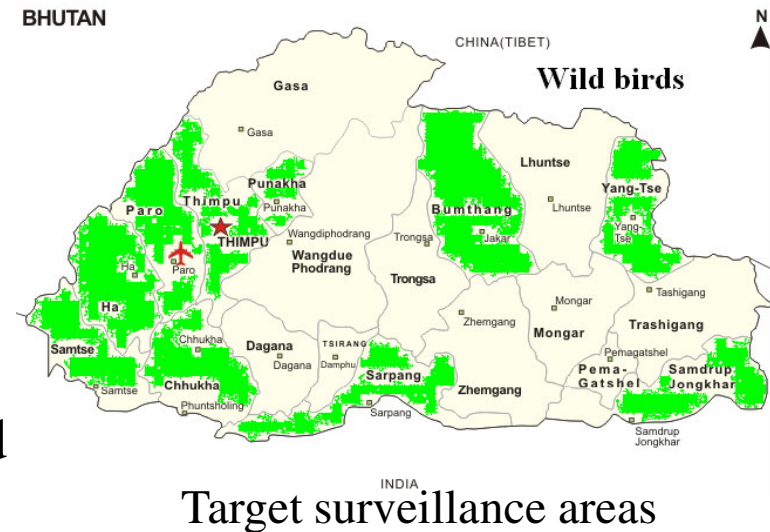


## Report-3: Continued.....



### 2. Coordination mechanism

- Any deaths in wild birds reported to NCAH through hotline
- Sampling/investigations coordinated through RLDC/SVLs/DVLs
- Regular sampling coordinated / planned with wildlife focal point
- Timing – October – March (intensified) and May, August
- High risk areas, along the rivers, wild bird roosting places, border areas
- Fresh fecal droppings collected in VTM





## Report-3: Continued.....



### 3. Results/Findings

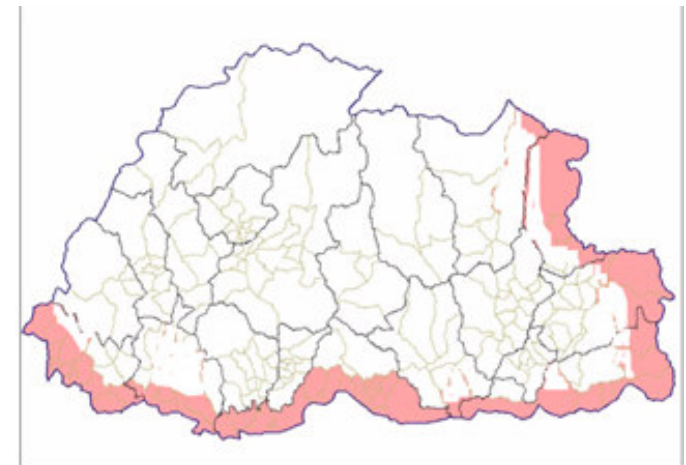
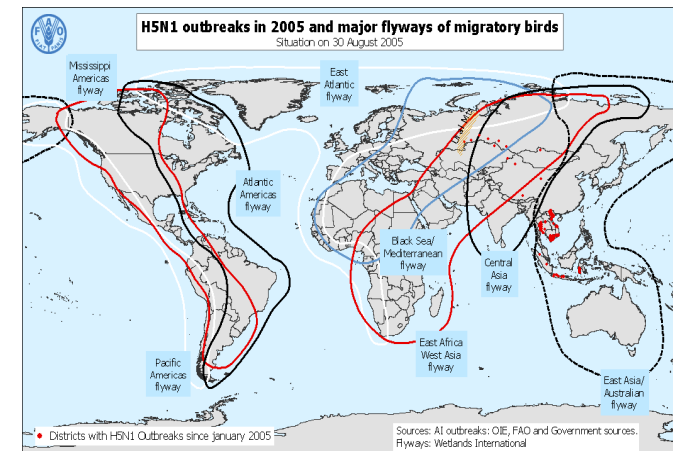
Year	Animal spp.	Sample type	Diagnosis	Remarks
		Feces /Droppings	rRT-PCR	
2010-2011	Wild birds	107	107	All negative
2011-2012	Wild birds	551	551	*All negative
*Except for a pigeon and a sparrow that tested positive from outbreak area				

- St Jude CEIRS collaboration on surveillance of wild & domestic birds, swine



## Implications of wild birds surveillance in the country

- Although Bhutan fall under Central Asian flyway, none of the wild birds during regular surveillance tested positive to H5N1 except for the sparrow and pigeon that tested positive for H5N1 from the outbreak area.
- Approximately 40 species of birds migrate to Bhutan from various places
  - Black necked cranes
  - Migratory water birds
- Frequent outbreaks of H5N1 virus in neighboring states
- Growing poultry industry in the country
- We believe in early detection through wild bird surveillance





## Plan ahead



- Strengthening Veterinary Vigilance & diagnostic facilities
  - Initiate virus isolation & Genetic analysis facilities at national level
  - Provide rRT-PCR diagnostic facilities at regional levels
- Widen surveillance in wild migratory and water birds (current focus is in high risk areas).
- Strengthen inter-sectoral collaboration in prevention and control
- Streamline education & awareness programs





**THANK YOU!**

The 5th OIE Regional Expert Group Meeting for Implementation of the Programme on Surveillance of Wild and Domestic Birds along Migratory Flyways,  
(Tokyo, Japan, 13-14 December 2012)

# *Avian Influenza Surveillance in Chinese Taipei*

*Shu-Fen, Chang*  
BAPHIQ

*The 5th OIE Regional Expert Group Meeting for Implementation of the  
Programme on Surveillance of Strengthening HPAI Control in Asia  
Tokyo, Japan, 13-14 December 2012*





# *Outline*

- Recent HPAI outbreaks in domestic birds
- Active AI surveillance programme of domestic birds
- Active/passive AI surveillance programme of wild birds





# Recent HPAI outbreaks in domestic birds

- No H5N1 case was noticed or detected in domestic and wild birds since the ancient times.
- H5N1 AIVs were only detected in smuggled birds (3 times in 2003, 2005 and 2012, respectively).
- 6 H5N2 subtype HPAI were detected in 2012.
  - Most of them were native broiler farms.
  - The predominated HPAIV is H5N2 with RRKR\*GLF in the HA0 cleavage site.

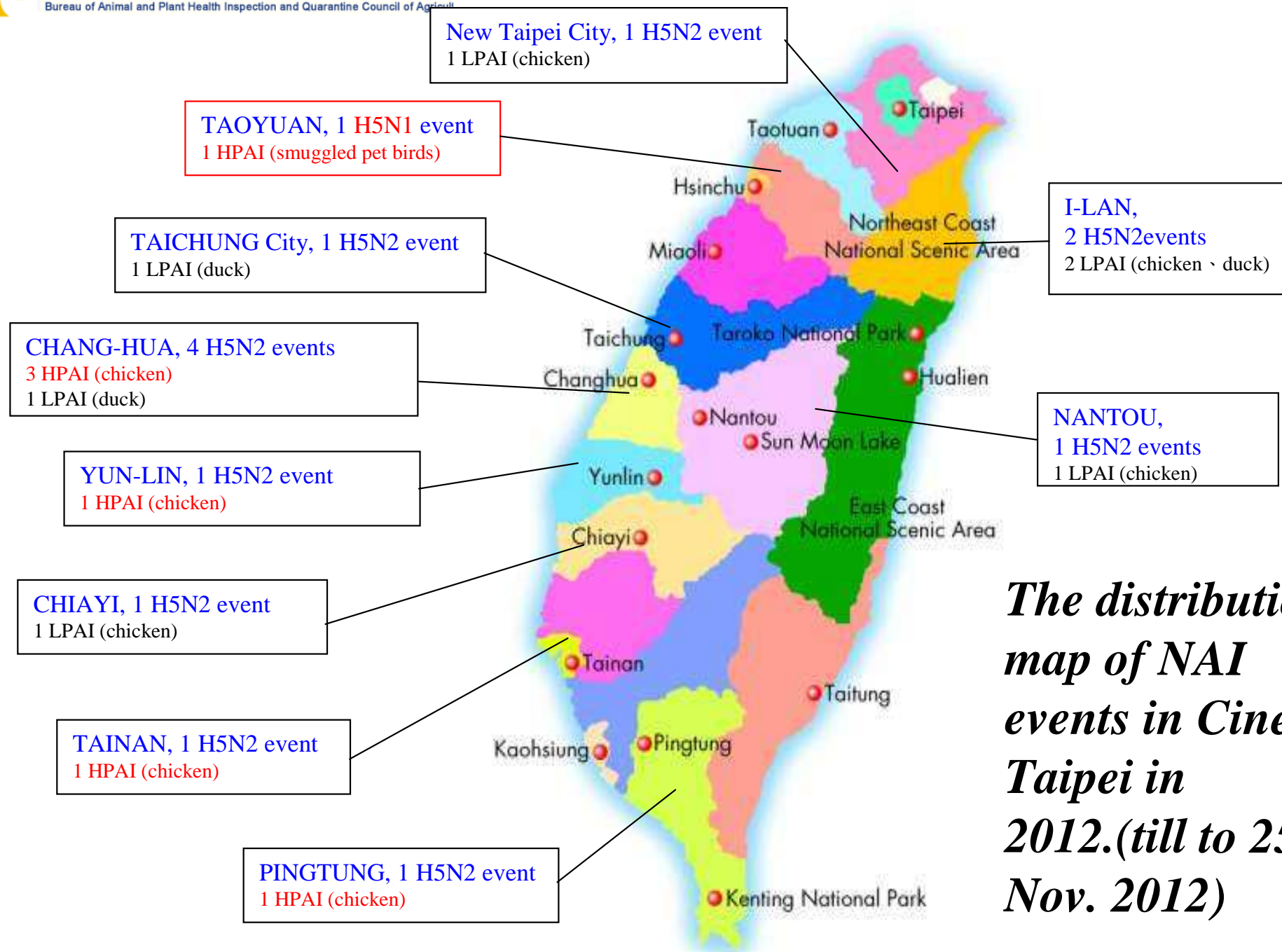
# Recent HPAI outbreaks in domestic birds

**Table 1. The detailed information of HPNAI cases in domestic birds in 2012**

	Event time	Species	Affected premises	Affected county	Virus characterization		
					Subtype	a.a. of HA0	IVPI
case1	Jan	Layer chicken	farm	Chang-Hua	H5N2	RKKR	2.01
case2	Feb	Broiler breeder	farm	Tai-Nan	H5N2	RRKR	2.53
case3	Feb	Native chicken	farm	Chang-Hua	H5N2	RKKR	2.28
case4	Mar	Layer chicken	farm	Chang-Hua	H5N2	RRKR	2.78
case5	Mar	Native chicken	abattoir	Ping-Tung	H5N2	RRKR	2.62
case6	May	Native chicken	farm	Yun-Lin	H5N2	RRKR	2.91

# Recent LPAI outbreaks in domestic birds

<b>Table 2. The detailed information of LPNAI cases in domestic birds in 2012</b>							
	Event time	Species	Affected premises	Affected county	Virus characterization		
					Subtype	a.a.sequence of HA0	IVPI
<b>case1</b>	Feb	Layer chicken	farm	Nan-Tou	H5N2	REKR	0.2
<b>case2</b>	Mar	Native	abattoir	I-Lan	H5N2	REKR	0.72
<b>case3</b>	Mar	Native chicken	market	New-Taipei	H5N2	REKR	-
<b>case4</b>	Mar	Meat duck	abattoir	I-Lan	H5N2	IETR	0.13
<b>case5</b>	Apr	Layer duck	farm	Chan-Hua	H5N2	IETR	0.00
<b>case6</b>	Apr	Meat duck	farm	Taichung	H5N2	IETR	-
<b>case7</b>	Nov	Layer chicken	farm	Chia-I	H5N2	REKR	-



*The distribution map of NAI events in Chinese Taipei in 2012.(till to 25 Nov. 2012)*



# Recent HPAI outbreak and H5N1 HPAI virus characterization in smuggled birds

- 38 smuggled pet birds delivered from Macao to Taiwan island were intercepted at the Airport on 7 July 2012.
- H5N1 HPAI virus had been identified from these birds. The result of HA0 cleavage site sequencing demonstrated high pathogenicity.
- The cleaning and disinfection of the environment and facilities were completed on 7 July 2012. All staff directly or indirectly contacting with the birds had been under monitoring by human health department for 10 days and being healthy at the duration.

**Table 3. The detailed information of HPAI case in smuggled pet birds in 2012**

Event time	Clinical signs	Species	Affected premises	Affected county	Virus characterization		
					Subtype	a.a. of HA0	IVPI
7/7	no	Japanese white-eye	airport	Tao-Yuan	H5N1	RERRRKR	-





# Active surveillance programme of domestic birds

- Active surveillance programme had been initiated since 1998.
- The aim is to detect clinical/subclinical NAI cases and as the measure for preacution for early case response and control.

Item	Description
Frequency	Four times a year
Timing	Each season
Target species	Chicken, Duck, Geese
Survey province	21 provinces
Sample category	Serum and Swab
Sample scale	20 /per farm
Target premise	Farms, markets
Target serotype	H5, H7
Testing method	ELISA, HI, RT-PCR, VI, Genetic analysis



# Active surveillance programme of domestic birds

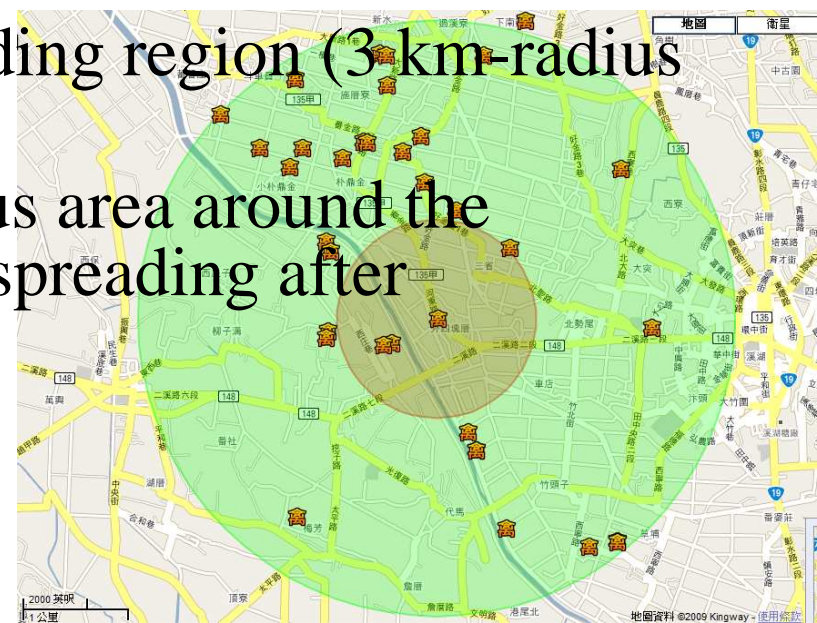
- All the HPAI cases in 2012 were detected by passive surveillance (reported cases).
- 5 out of 7 LPAI cases in 2012 were detected by active surveillance or intensified surveillance on surrounding poultry farms of the infected farms.

Table 4. Number of sampling poultry farms and positive farms detected by active AI surveillance			
	2010	2011	2012 (Jan-Oct)
Chicken farm	776	658	642
Duck farm	331	283	230
Goose farm	82	79	72
NAI positive farm	2 (0.17%)	2 (0.2%)	5 (0.5%)

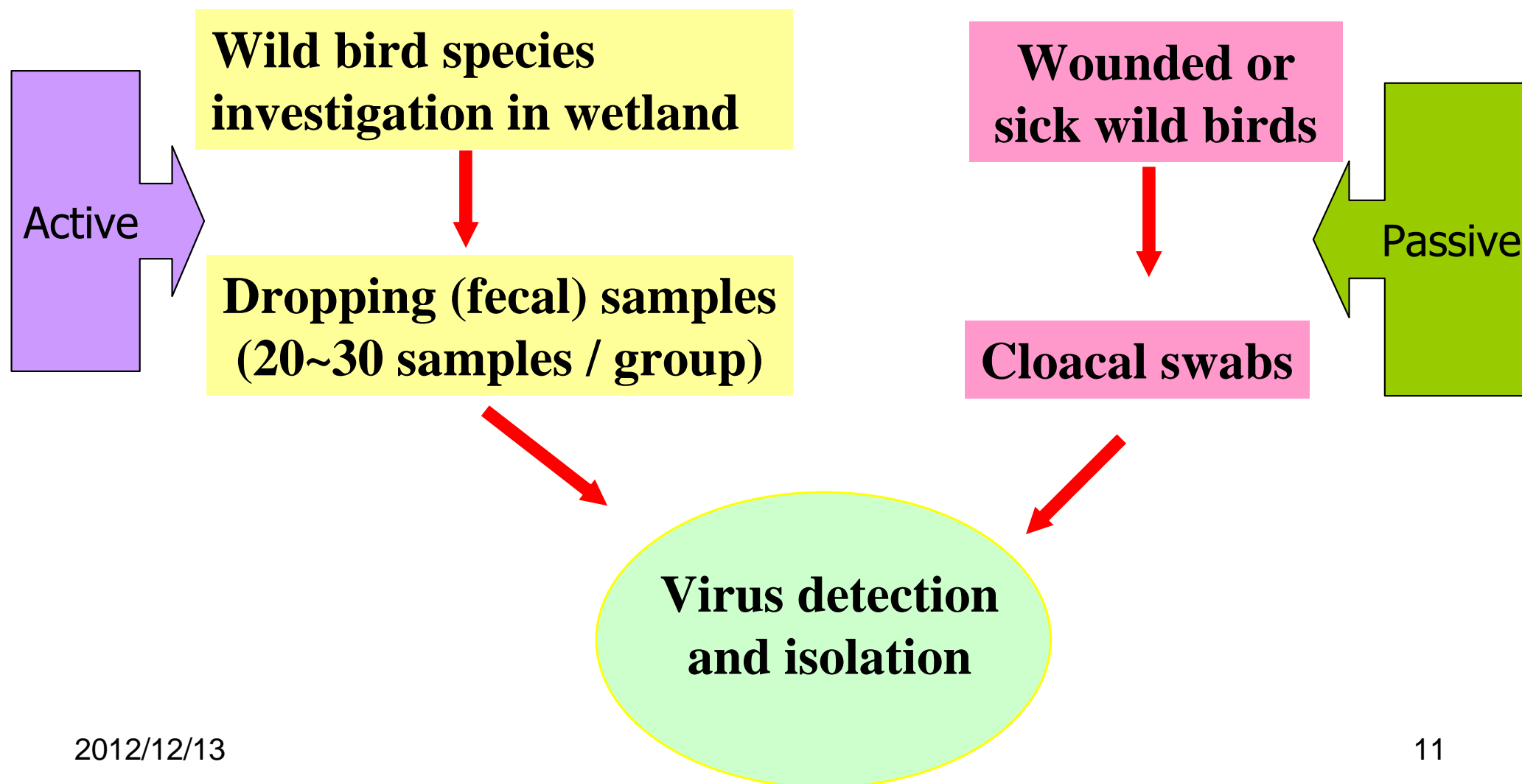


# Active surveillance programme of domestic birds

- Control measures to be undertaken if HPAI case is detected:
  - Movement restriction.
  - Stamping-out of infected farm.
  - Cleaning and disinfection.
  - Empty for at least 21 days.
  - Sentinel chicken testing.
  - Intensified surveillance of surrounding region (3 km-radius of infected farm) for 3 months.
  - Ring vaccination within 1 km radius area around the infected farm (if the outbreak was spreading after evaluation).



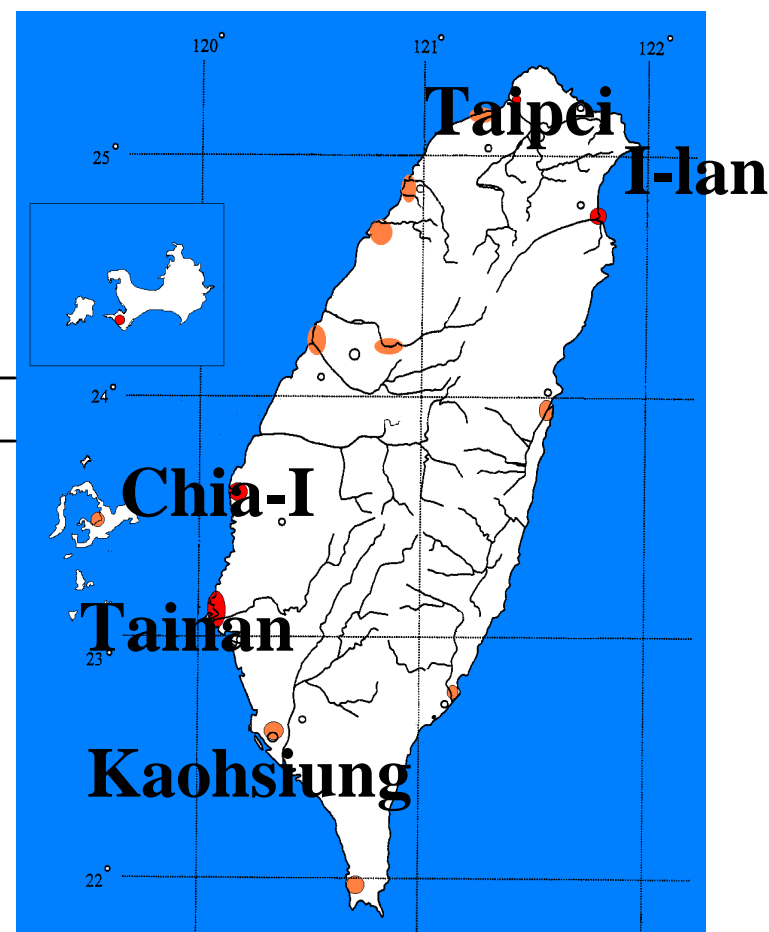
# Active/passive surveillance programme of wild birds (WB)



# Active surveillance programme of WB

- Dropping samples collected from wetlands
- >3,000 samples were tested annually by RT-PCR and VI
- Samples were tested negative for HPAIV from 1998 to 2012

Item	Description
Frequency	Monthly
Timing of sampling	Each season, intensified surveillance on fall and winter
Target species	Ducks (Anatidae), Shorebirds or others
Sample category	Droppings (swab)
Sample scale	20-30 samples / group
Target serotype of influenza A	H5, H7
Testing method	RT-PCR, VI, Genetic analysis





# Active surveillance programme on WB

Year	Ducks	Shorebirds	Gulls	Egrettas	Others	Total
1998	1,457	0	0	30	0	1,487
1999	1,418	75	0	0	0	1,493
2000	1,825	44	0	0	0	1,869
2001	2,516	<b>45</b>	3	0	0	2,564
2002	2,060	652	72	0	98	2,882
2003	1,831	553	59	0	0	2,443
2004	1,902	1,149	190	4	106	3,351
2005	2,806	1,357	112	38	194	4,507
2006	2,352	1,413	161	436	179	4,541
2007	2,645	1,147	20	317	21	4,150
2008	2,768	920	80	389	108	4,265
2009	2,755	835	57	417	91	4,155
2010	2,791	754	79	504	18	4,246
2011	2,301	1,068	40	326	26	<b>3,761</b>
<b>Sum</b>	<b>31,427</b>	<b>10,034</b>	<b>873</b>	<b>2,461</b>	<b>841</b>	<b>45,714</b>
<b>isolates</b>	<b>340</b>	<b>14</b>	<b>2</b>	<b>6</b>	<b>1</b>	<b>363</b>
Prevalence(%)	1.08	0.14	0.23	0.24	0.12	0.80

<i>Detected subtypes (46)</i>
H1N1, H1N2, H1N3, H2N3, H2N7, H2N9, H3N2, H3N6, H3N7, H3N8, H3N9, H4N2, H4N3, H4N5, H4N6, H4N7, H4N8, <b>H5N2, H5N6,</b> H6N1, H6N2, H6N5, H6N9, <u>H7N1, H7N2, H7N3, H7N5, H7N6, H7N7, H7N9,</u> H8N3, H8N4, H9N6, H9N9, H10N1, H10N2, H10N3, H10N4, H10N6, H10N7, H10N8, H10N9, H11N3, H11N9, H12N2, H14N7

All isolates are LPAIV

Subtype	Cleavage site of HA protein
H7	PEIPKGR*GLF
H5	PQRETR*GLF



# Active surveillance programme on WB

## Subtypes of AIVs isolated from WB (1998-2011)

Subtype	N1	N2	N3	N4	N5	N6	N7	N8	N9	Total
H1	12	1	5							18
H2			9				2		1	12
H3		2				12	1	32	1	48
H4		8	2		3	125	4	9		151
H5		6				1				7
H6	7	3			1				1	12
H7	9	3	13		2	5	4		8	44
H8			1	2						3
H9						1			1	2
H10	1	1	19	8		3	12	5	4	49
H11			2						8	10
H12		1								1
H13										0
H14							2			2
H15										0
H16										0
Total	29	25	51	10	6	147	25	46	24	363



# Passive surveillance programme on WB

Table 5. Results of passive surveillance in WB

Year	Waterfowls (AIV isolates)	Land birds (AIV isolates)	Total (AIV isolates)
1998-2007	70 (0)	7 (1*)	77 (1)
2008	15 (0)	75 (0)	90 (0)
2009	267 (0)	625 (0)	892 (0)
2010	58 (0)	555 (0)	613 (0)
2011	2 (0)	108 (0)	110 (0)
Total	412 (0)	1,370 (1)	1,782 (1)

1. Samples were from dead, wounded or sick wild birds

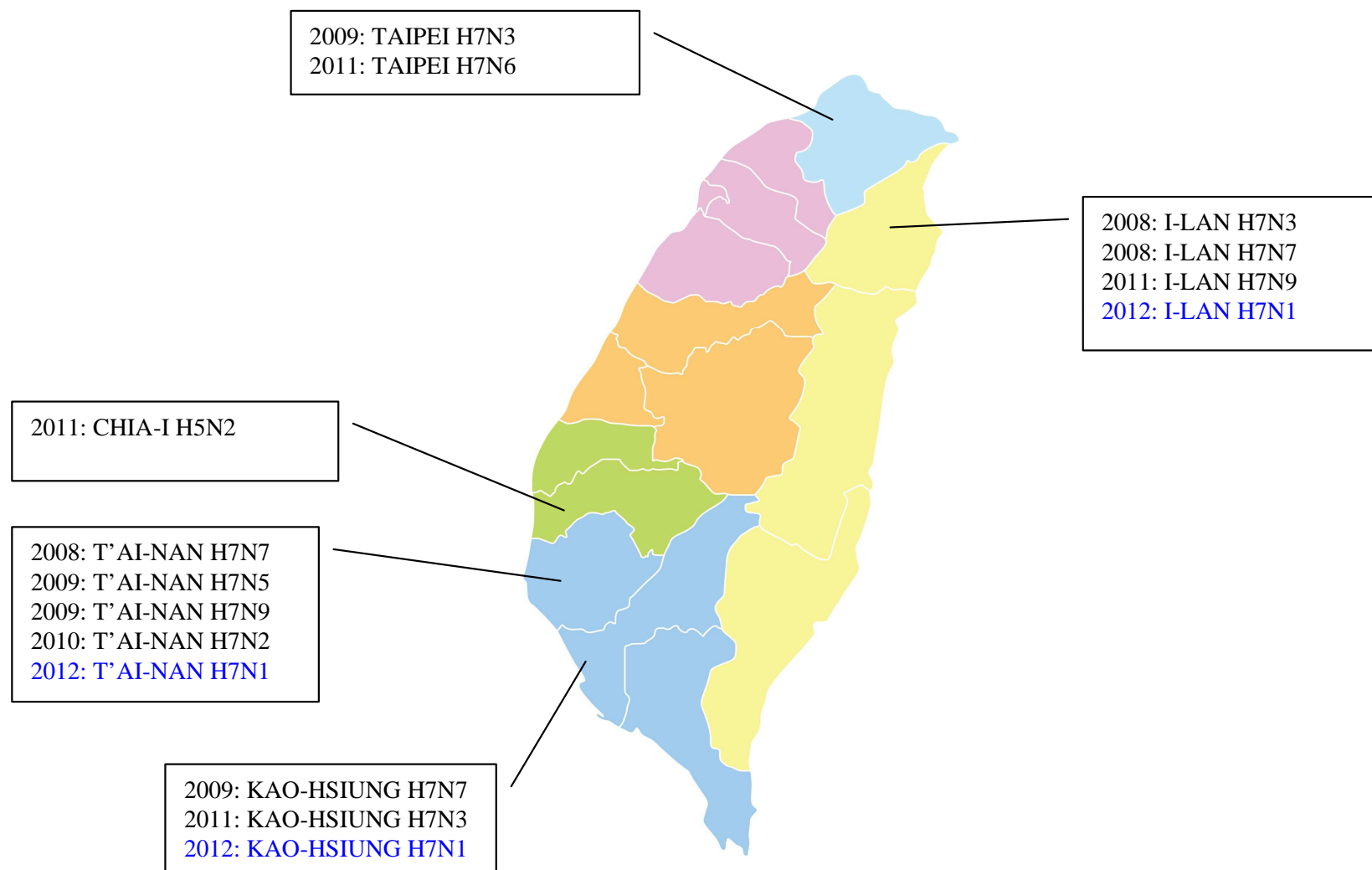
2. \* Crested serpent-eagle/Hualien/A423/05(H12N2)







# The distribution map of H5/H7 AIVs isolated from WB from 2008 to 2012





# Overall observations

- *Passive surveillance is more efficient in detect HPAI cases in the field, and active surveillance system could usefully detect the clinical or subclinical NAI cases in the domestic and wild bird population. All of them are needed to declare or maintain the disease free situation on the scientific basis.*
- We could detect the NAIIV, such as H7N3 and H5N2, from the drooping samples of migratory birds before NAI with same subtype outbreak in poultry. Even though there is no close relation between viruses isolated from wild bird droppings and poultry swabs, we believe that the migratory birds indeed play a role for NAI introduction according to the experience of disease countries, but the mechanism how migratory birds introduce/ transmit/ spread the AIV is still unclear.

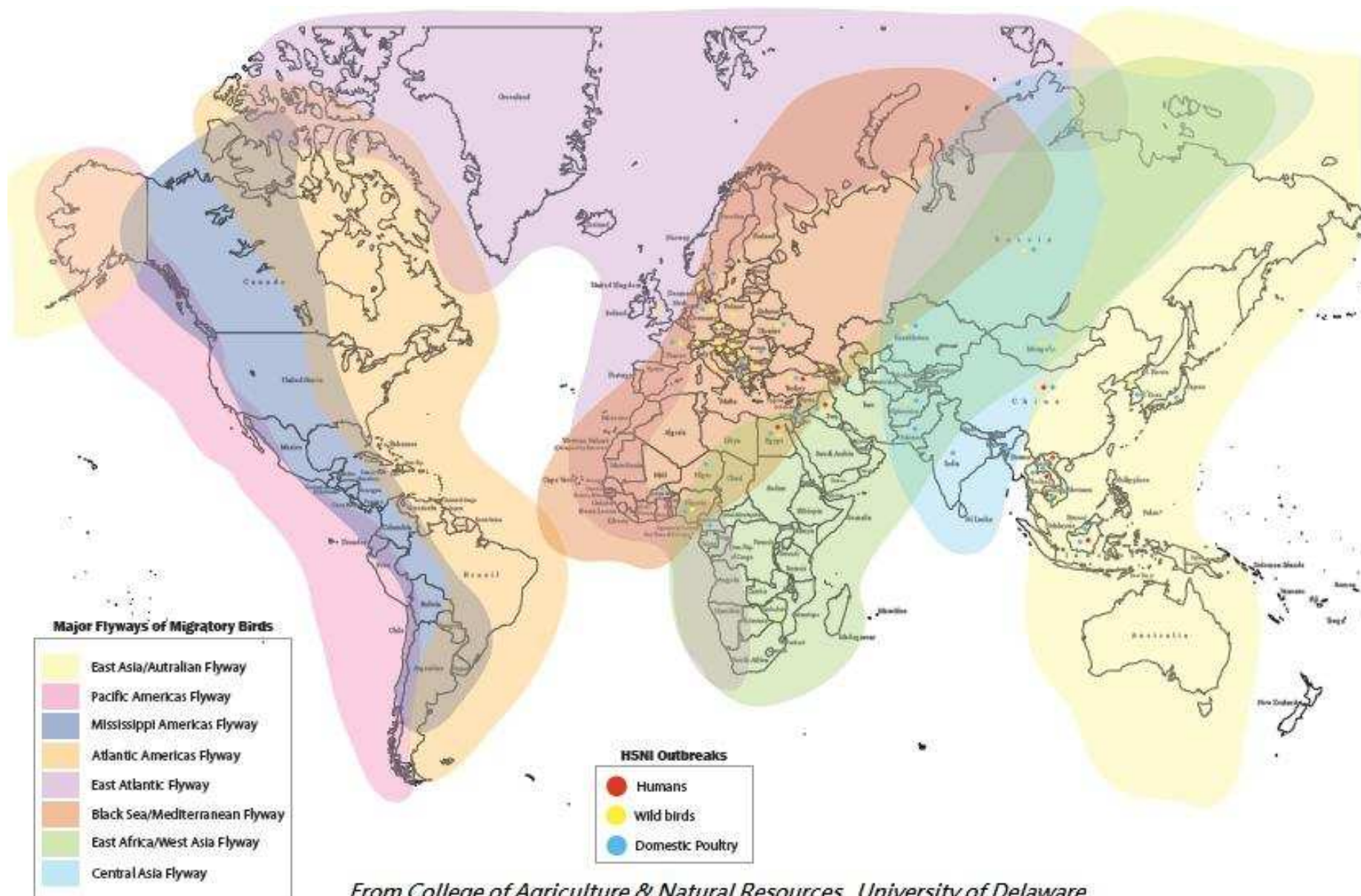


# Plan for 2013 forward

- Continuously conducting active/ passive AI surveillance in domestic and wild birds.
- Participating the OIE regional cooperation for HPAI prevention and control.
- Revising the response manual for NAI case control according to what we learnt from the outbreaks and information got from surveillance as well as revised OIE related articles.

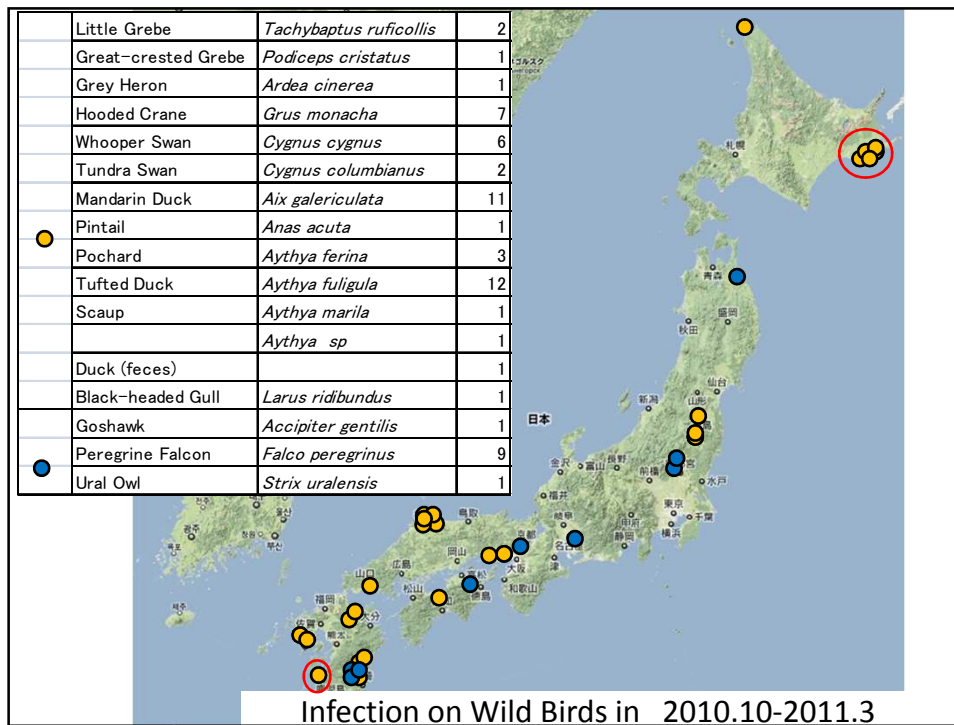


# *Thank you for your attention!*



*From College of Agriculture & Natural Resources, University of Delaware*





## Izumi, Kagoshima

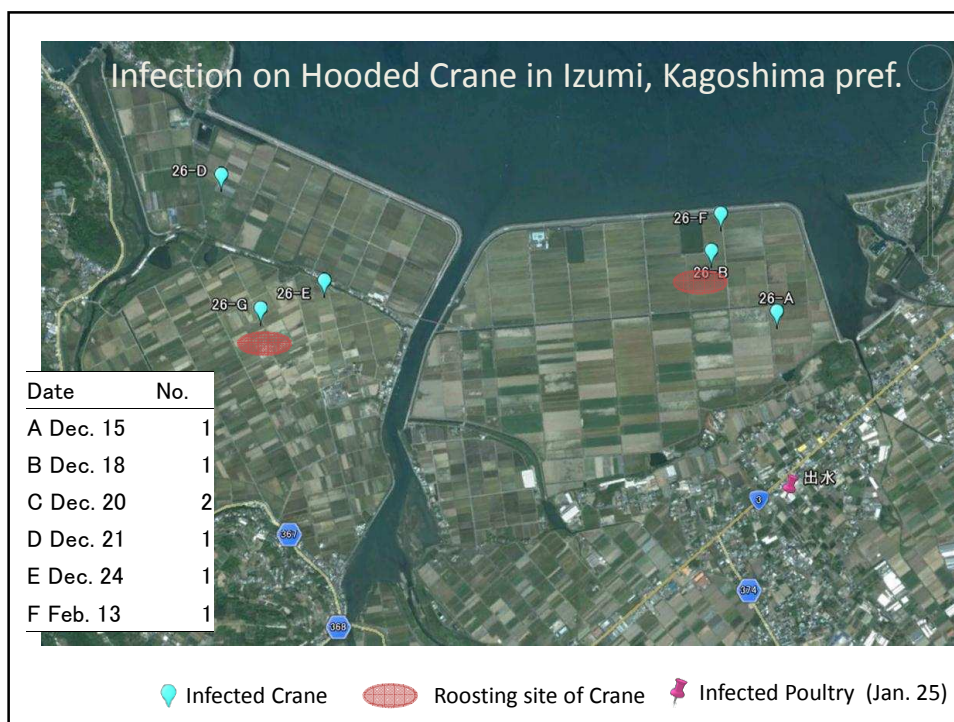
Wintering number of Crane

: Hooded Crane *Grus monacha*:10,000

White-naped Crane *Grus vipio* : 3,000



With many ducks and poultry farms



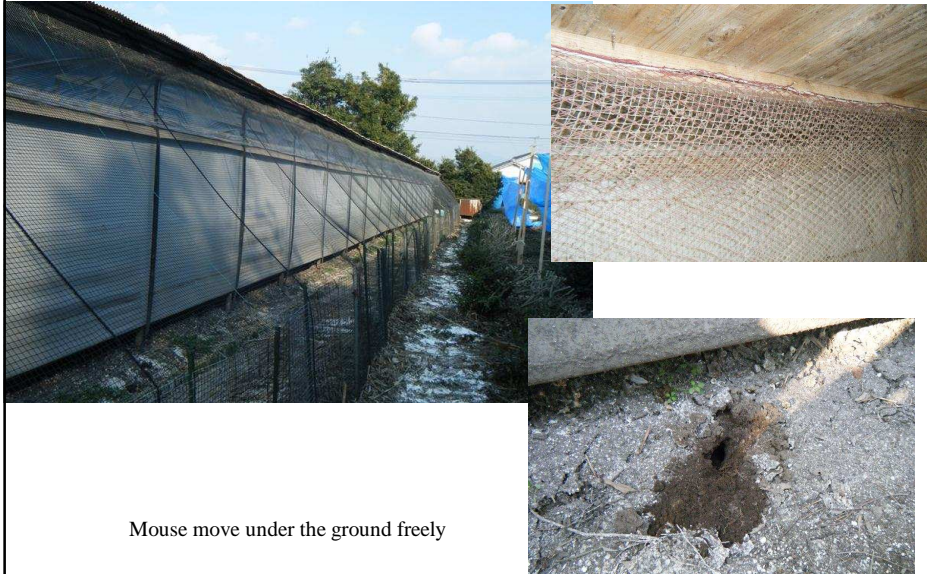
## Prevention of epidemics in Izumi City

1. Monitoring of infected Birds
2. Close facilities for tourist
3. Keep off the main habitat of cranes
4. Disinfect cars at the boundary of keep off area



### Infection farm at Izumi

Bird net covered all house, outside and inside



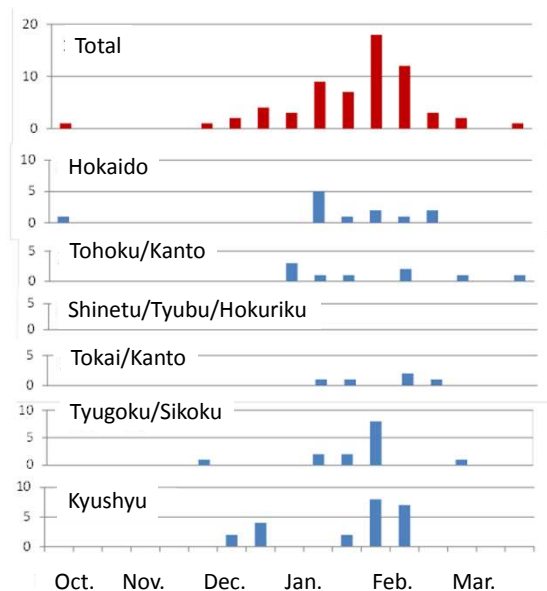
Mouse move under the ground freely

### Dusky Thrush *Turdus naumanni* in infected farm, Izumi





## Time of infection



## Area

		Hokkaido	Tohoku Kanto	Shinetsu Tyubu Hokuriku	Tokai Kinki	Tyugoku Sikoku	Kyushyu	Total
Little Grebe	<i>Tachybaptus ruficollis</i>				1		1	2
Great-crested Grebe	<i>Podiceps cristatus</i>				1			1
Grey Heron	<i>Ardea cinerea</i>						1	1
Hooded Crane	<i>Grus monacha</i>						7	7
Whooper Swan	<i>Cygnus cygnus</i>	7						7
Tundra Swan	<i>Cygnus columbianus</i>		1			1		2
Duck (feces)		1						1
Mandarin Duck	<i>Aix galericulata</i>					1	10	11
Pintail	<i>Anas acuta</i>	1						1
Pochard	<i>Aythya ferina</i>				1	2		3
Tufted Duck	<i>Aythya fuligula</i>		5			7		12
Scaup	<i>Aythya marila</i>	1						1
	<i>Aythya sp</i>	1						1
Black-headed Gull	<i>Larus ridibundus</i>					1		1
Goshawk	<i>Accipiter gentilis</i>		1					1
Peregrine Falcon	<i>Falco peregrinus</i>		2		2	1	4	9
Ural Owl	<i>Strix uralensis</i>					1		1
Total		12	9		5	14	23	62
Zoo				1		1		2
Poultry farm			2		6	1	17	24

## Environment of Infection site

	Near sea	Lake near sea	Inland lake	Inland river	City	Rural area	Mountain	Total
Hokkaido	1	4						5
Tohoku/Kanto		1	1	1		2		5
Shinetsu/Tyubu/Hokuriku								
Tokai/Kinki	1		1		1	1		4
Tyugoku/Sikoku		4					2	6
Kyushu	4	2		1		3	3	13
Total	6	11	2	2	1	6	5	33

## Pick up point of samples

	Water fowl	Raptors	Total
Water area	20	–	20
Land	29	11	40
Unknown	2	–	2
Total	52	11	62

## Conclusion

- There are high possibility that HPAI virus came into japan with migratory waterfowl in autumn.
- HPAI virus were kept in waterfowl flocks and increased at late January.
- The density of HPAI virus in western Japan higher than eastern Japan where the large number of ducks wintered.
- It need to pay attention to movement of wintering bird in the wintering area.
- Monitoring and prevention of HPAI virus in East Asia are important.