

WOAH standards related to VBDs: animal and vector surveillance



WOAH Regional Workshop on
Vector Borne diseases in Asia
and the Pacific

19-20 September 2024

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World
Organisation
for Animal
Health
Founded as OIE

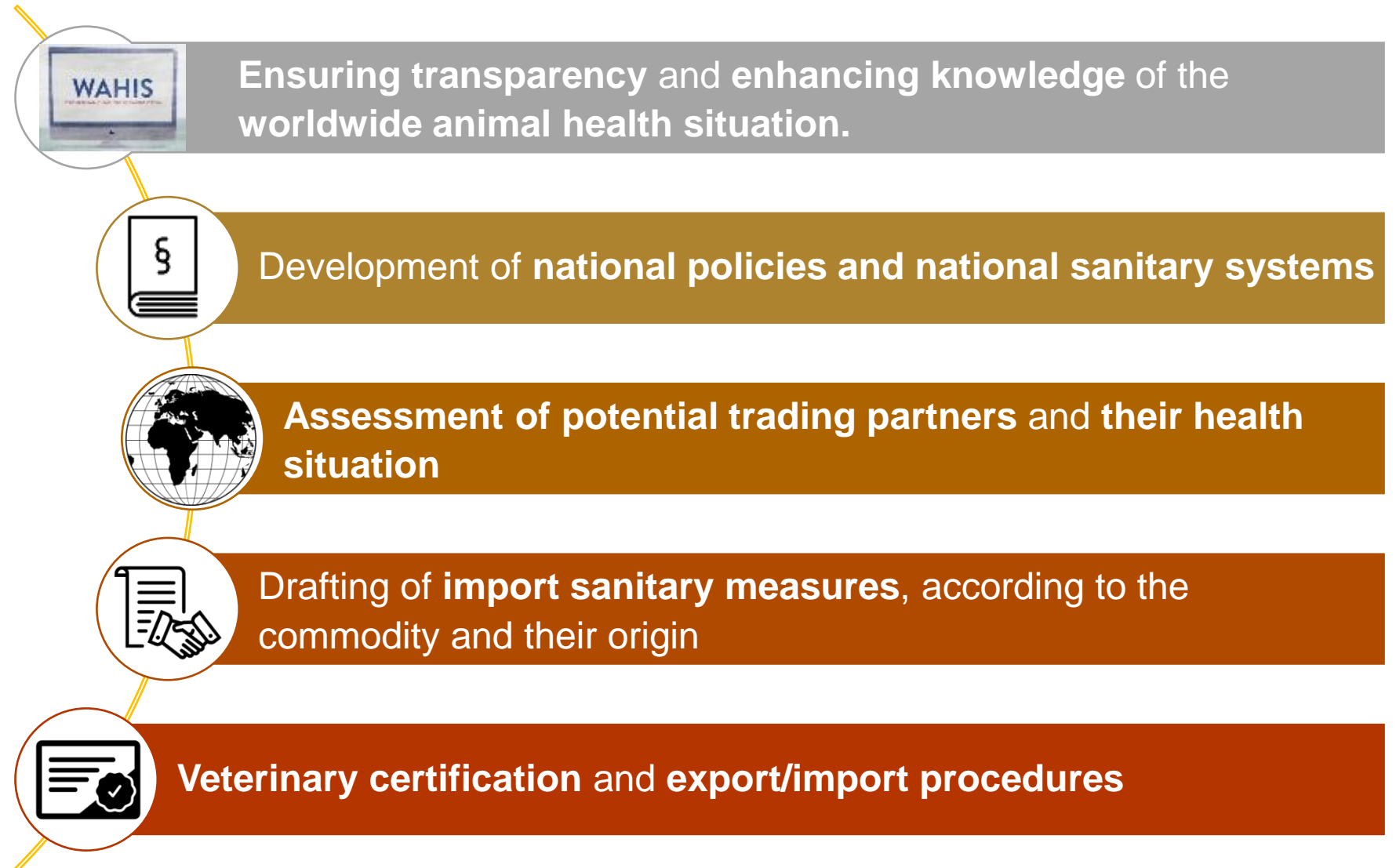
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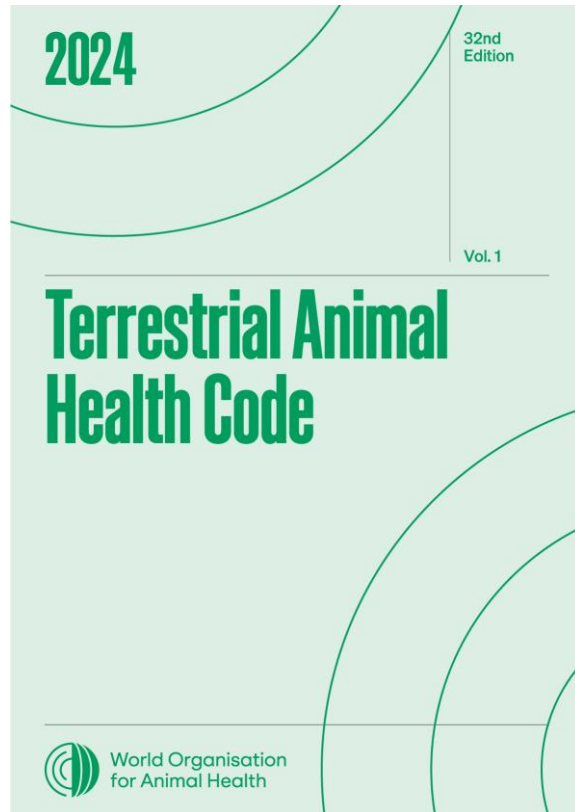


WOAH international standards

WOAH establishes standards for the improvement of animal health and welfare and veterinary public health worldwide, including the prevention of disease spread through international trade of animals and animal products.

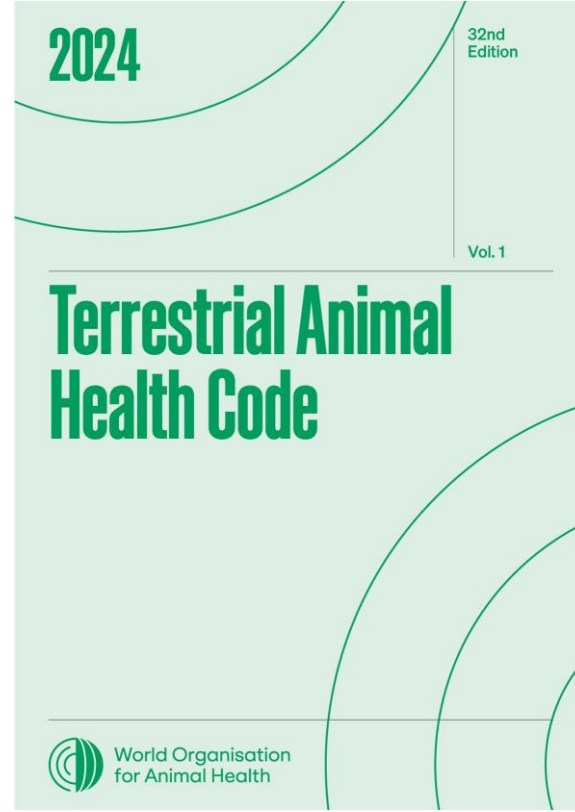


Terrestrial Code



Volume I

User's guide and
Horizontal chapters



Volume II

Disease-specific
chapters

- New version every year
- Latest version publicly available on the WOAH website
- Previous editions are also available





Terrestrial Code - content

Volume I: Horizontal chapters

- User's Guide
- Glossary
- Animal disease diagnosis, surveillance and notification
- Risk analysis
- Quality of Veterinary Services
- Disease prevention and control
- Trade measures, import-export procedures and veterinary certification
- Veterinary public health
- Animal welfare

Volume II: Disease-specific chapters

- **Definition of infection/ infestation** and specific epidemiological considerations
- **Safe commodities** where appropriate
 - Safe commodities = based on absence of the pathogenic agent in the traded commodity OR inactivation by processing or treatment that the commodity has undergone
 - Importing countries should not apply trade restrictions to safe commodities with respect to the pathogenic agent concerned
- **Determination of the animal health status** of a country, zone or compartment
 - Official status recognition
 - Self declaration of animal health status
- **Recommendations on safe trade** for live animals, genetic material, other products of animal origin (meat, milk, eggs, skins, etc.)
- **Specific management** of commodities, e.g.
 - Recommendations on inactivation
 - Vector-protection during transport
- **Recommendations on surveillance**
 - Surveillance strategies specific to infection/ infestation
 - Link to Chapter 1.4 and 1.5 on animal health and vector surveillance



Glossary

- **Vector**

means an insect or any living carrier that transports an infectious agent from an infected individual to a susceptible individual or its food or immediate surroundings. The organism may or may not pass through a development cycle within the vector.

- **Infestation**

means the external invasion or colonisation of animals or their immediate surroundings by arthropods, which may cause clinical signs or are potential vectors of pathogenic agents.

- **Official control programme**

means a programme which is approved, and managed or supervised by the Veterinary Authority of a Member Country for the purposes of controlling a vector, pathogenic agent or disease by specific measures applied throughout that Member Country, or within a zone or compartment of that Member Country.



SECTION 1. ANIMAL DISEASE DIAGNOSIS, SURVEILLANCE AND NOTIFICATION

- Chapter 1.1.** Notification of diseases and provision of epidemiological information
- Chapter 1.2.** Criteria for the inclusion in the OIE list
- Chapter 1.3.** Diseases, infections and infestations listed by the OIE
- Chapter 1.4.** Animal health surveillance
- Chapter 1.5.** Surveillance for arthropod vectors of animal diseases

SECTION 4. DISEASE PREVENTION AND CONTROL

- Chapter 4.4.** Zoning and compartmentalisation
- Chapter 4.5.** Application of compartmentalisation
- Chapter 4.18.** Official control programmes for listed and emerging diseases

A photograph of a dirt road winding through a lush tropical forest. In the foreground, two people are walking away from the camera, each carrying a large, heavy load of sticks or branches balanced on their heads. The person on the left is wearing a blue shirt and a patterned skirt, while the person on the right is wearing a light-colored shirt and dark pants. The road is flanked by dense green vegetation, including various types of palm trees and other tropical plants. The sky is overcast and grey.

Specific WOAAH Standards

**Terrestrial Code
Chapter 1.5.
Surveillance for arthropod
vectors of animal diseases**



Chapter 1.5. Article 1.5.1. Introduction

- The Terrestrial Code contains recommendations for the surveillance of several vector-borne diseases and general recommendations for animal health surveillance.
- The need has arisen to complement these general recommendations on surveillance with advice on the surveillance for vectors themselves. This chapter only addresses surveillance for arthropod vectors.
- For the **purpose of trade**, it should be noted that there is no conclusive relationship between the presence of vectors and the disease status of a country/zone, and also that **the apparent absence of vectors does not by itself confirm vector-free status**.



First adopted in 2009;

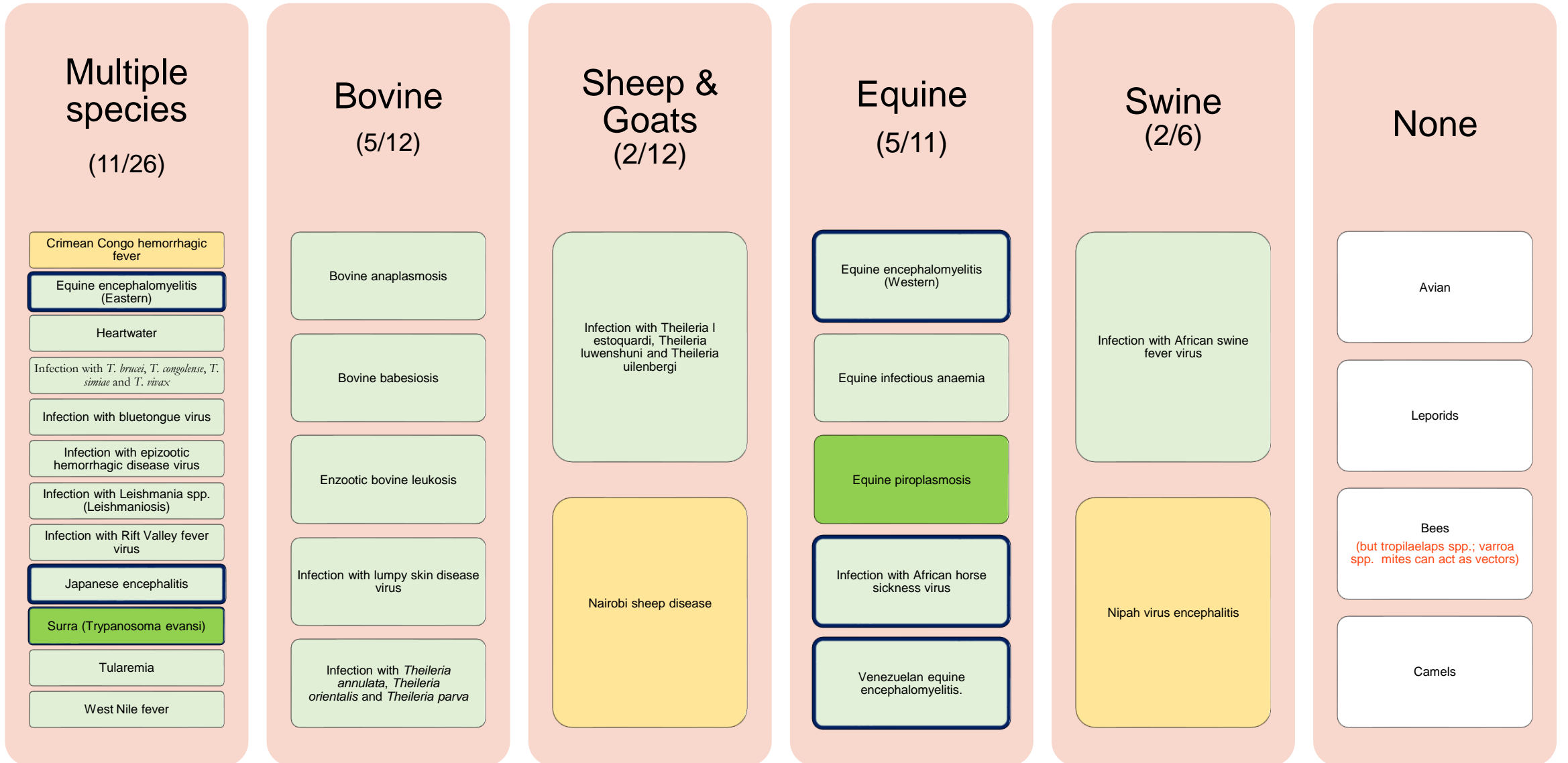
Most recent update adopted in 2010.



Chapter 1.5. **Article 1.5.2. Objectives**

- 1) **gathering up-to-date information** on the spatial and temporal distribution and abundance of vectors of the arthropod-borne listed diseases and emerging diseases;
- 2) **monitoring changes** in the spatial and temporal distribution and abundance of these vectors;
- 3) collecting relevant data to **inform risk assessment** (including vector competency) and risk management of these vector-borne diseases;
- 4) **detecting the presence of specific vectors or confirming their absence**;
- 5) understanding **pathways of entry** for vectors and vector-borne pathogenic agents.

Article 1.5.3. Sampling methodology: Sampling methods- Data management, analysis and interpretation



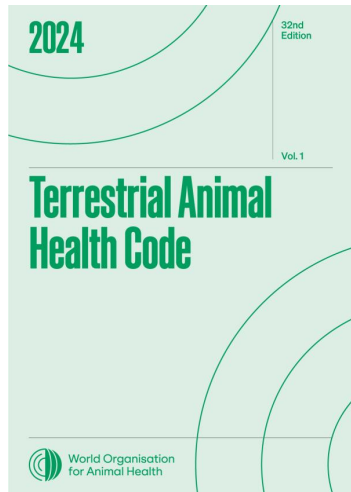
Adopted in 2024-2023

Terrestrial Code Chapter exists

Chapter under revision

In preparation

Volume II: Disease-specific chapters



1. Definition of infection/ infestation, its occurrence and specific epidemiological considerations (including relevant vectors)
2. Determination of the animal health status of a country, zone or compartment
3. Recommendations on safe trade for live animals, genetic material, other products of animal origin (meat, milk, eggs, skins, etc.)
4. Recommendations on surveillance

Terrestrial Manual



Complementary Chapters – Regularly updated

1. Standards for diagnosis and vaccines
2. Epidemiological information
3. Detailed information on vectors, vector species, etc

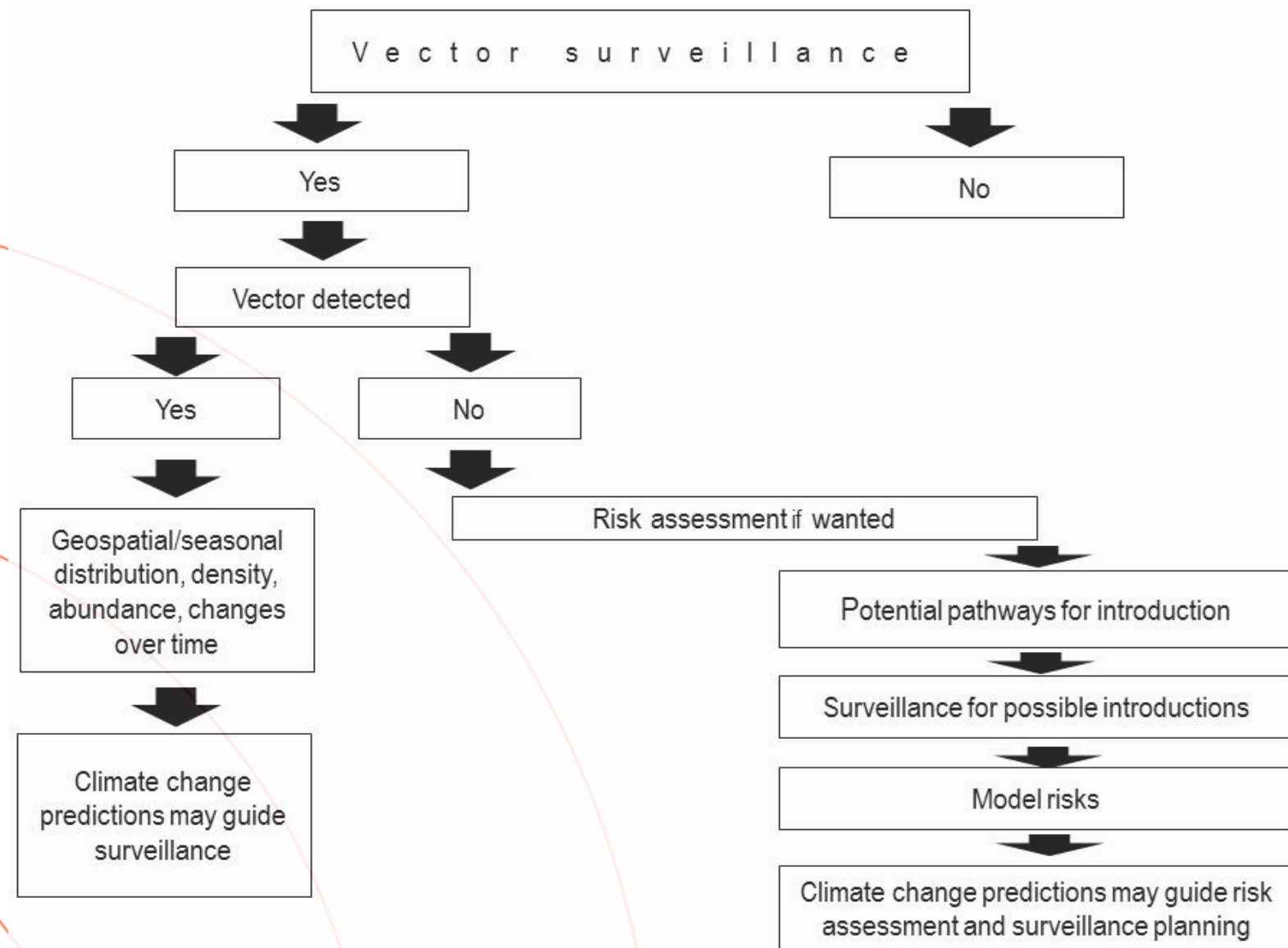


Specific considerations for VBD

1. **Definition of disease occurrence**
 - Detection of the pathogenic agent in an animal (not vector)
2. **Animal health Status**
 - Vector Surveillance – presence/absence of vector
 - Seasonal Freedom
 - High surveillance area bordering infected country or zone
3. **Protection of animals from vectors**
 - vector-protected establishment or facility
 - During transport
4. **Vector related sanitary measures for trade**
 - Free of ticks - treatment
 - Protection from vector attacks during transport/transit



Chapter 1.5.





Using Terrestrial Code Standards on Vector Surveillance

Chapter 1.4. Animal health surveillance

Chapter 1.5. Surveillance for arthropod vectors of animal diseases

Chapter 4.4. Zoning and compartmentalisation

Chapter 4.18. Vaccination

Chapter 4.19. Official control programmes for listed and emerging diseases

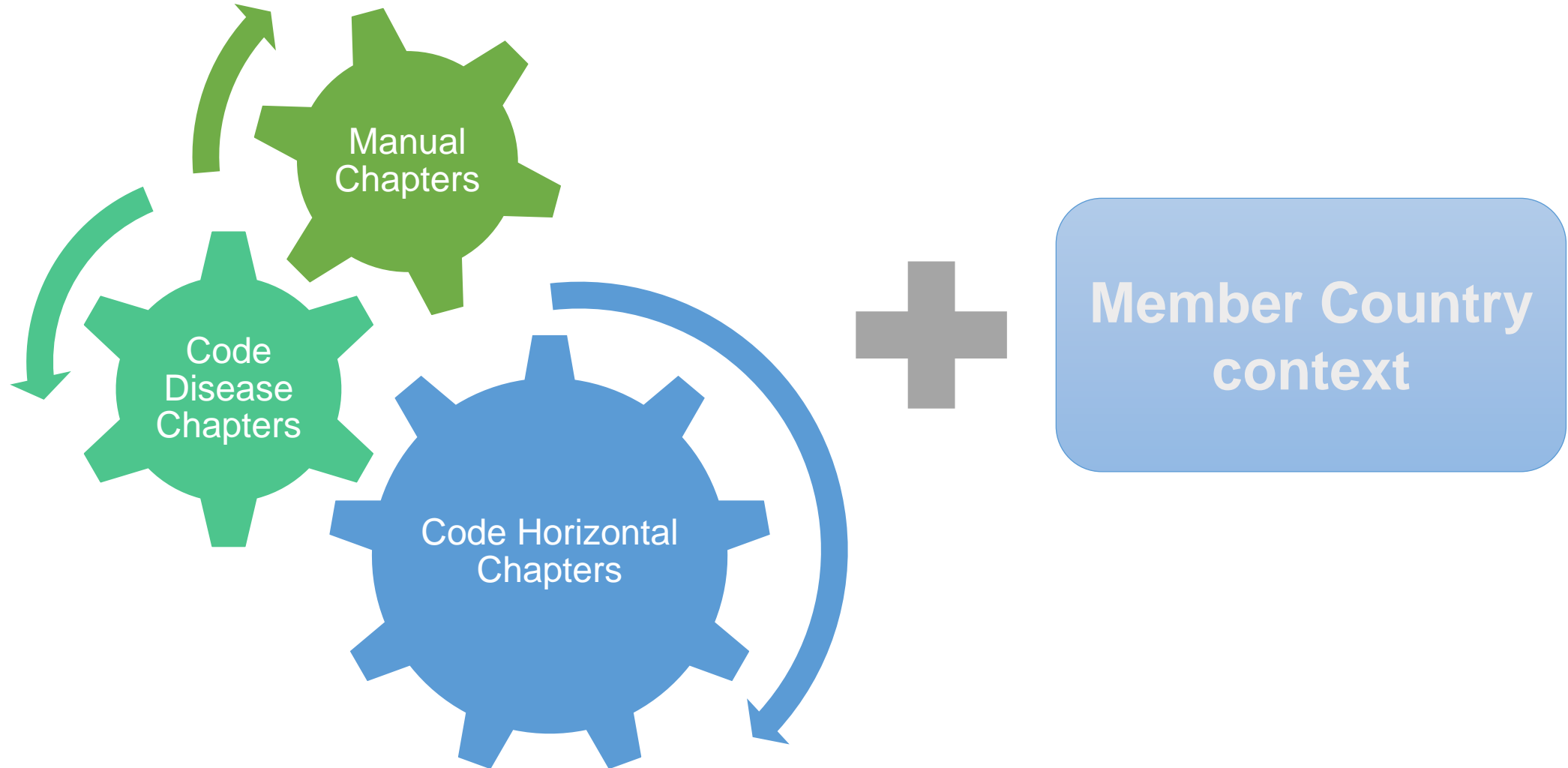
Volume II: Disease-specific chapters

Chapter 1.6. Procedures for official recognition of AH status, by WOA

Chapter 1.7.-1.12. Application for official recognition by WOA of free status for ...



Using the WOAAH international standards



Thank you

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WOAH Regional Workshop on Vector Borne diseases in Asia and the Pacific

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Updates on recent trends of VBDs Globally and in Asia Pacific reported to WOAH (2023-2024)

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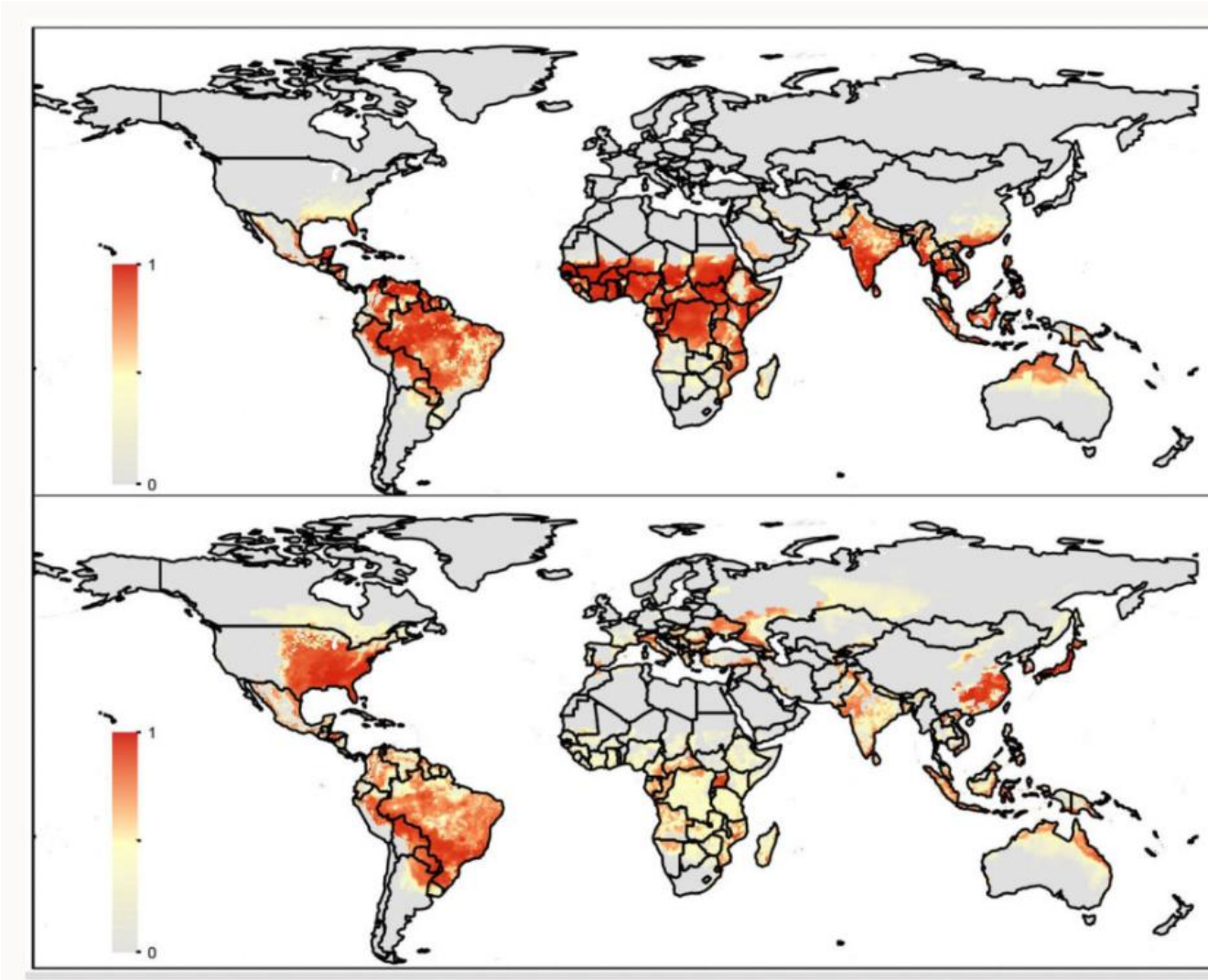
Paolo Tizzani
Veterinary epidemiologist
Data Integration Department



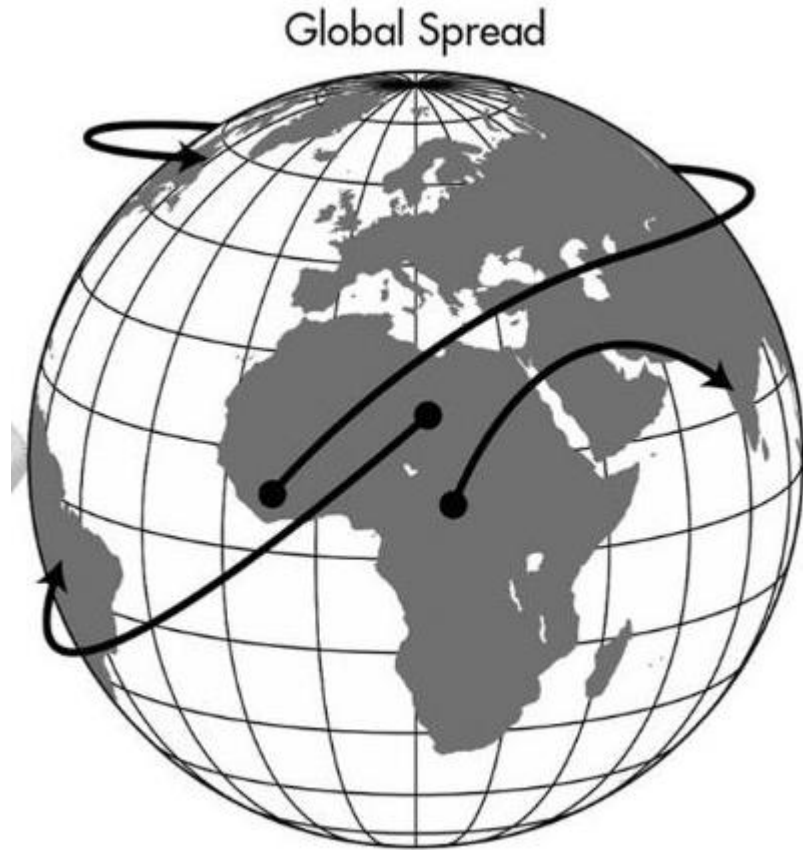
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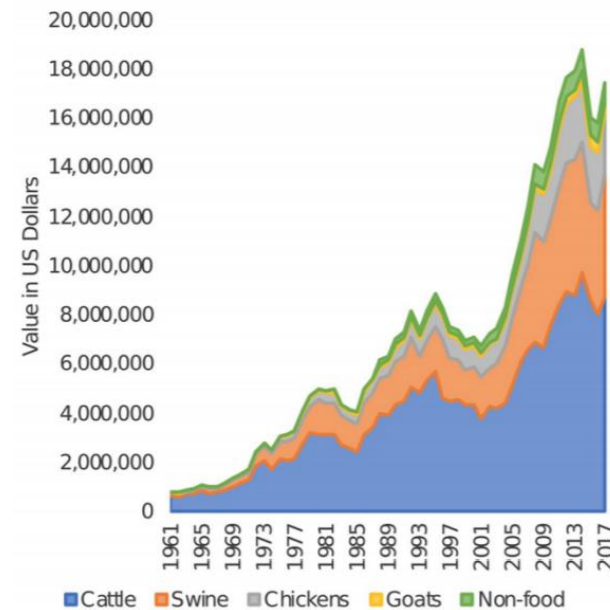
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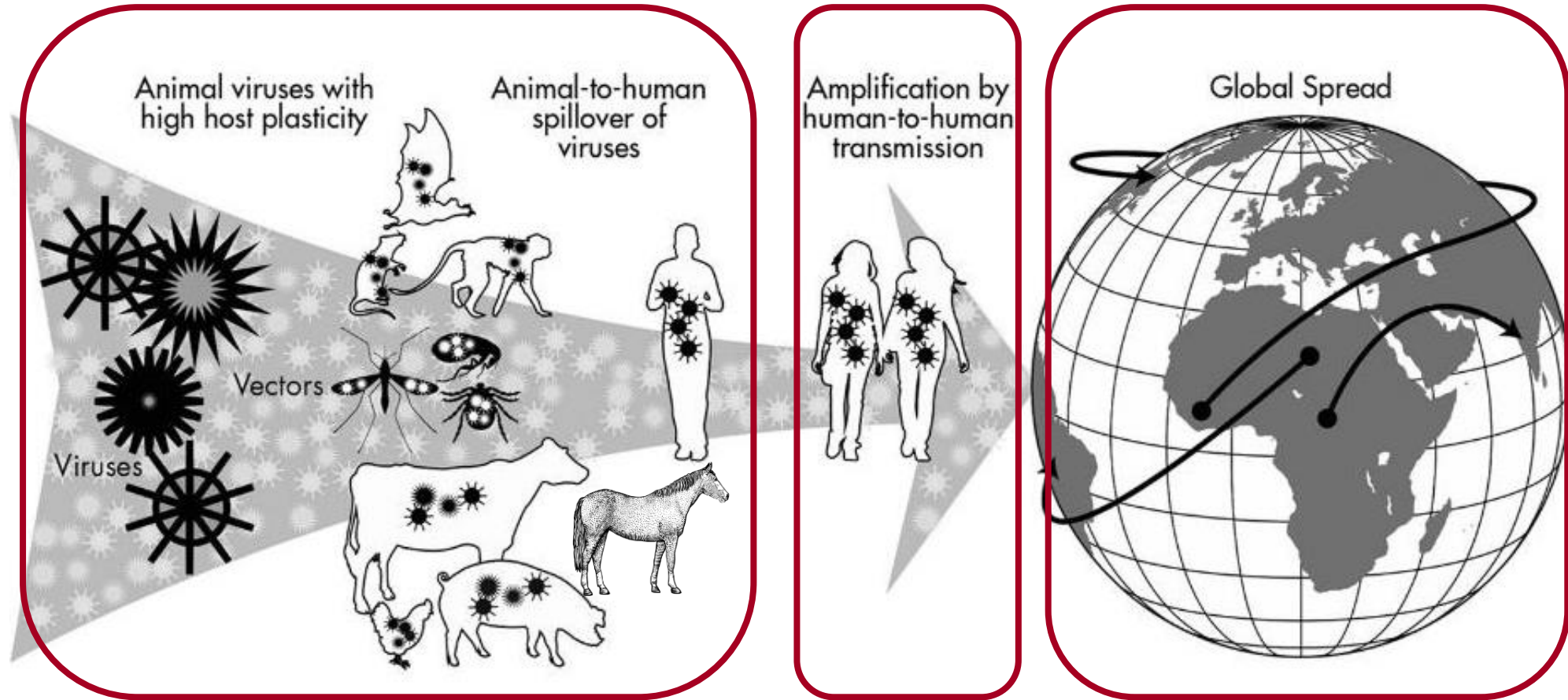
These maps show the predicted global ranges of *Aedes aegypti* (above) and *Aedes albopictus* (below) in 2050 assuming a 'medium' climate scenario in which greenhouse gas emissions peak in 2080 and then begin to decline. The darker areas have the highest predicted prevalence of mosquitoes. MORITZ KRAEMER FOR NATURE MICROBIOLOGY



+11 million passengers fly everyday (IATA, 2020)



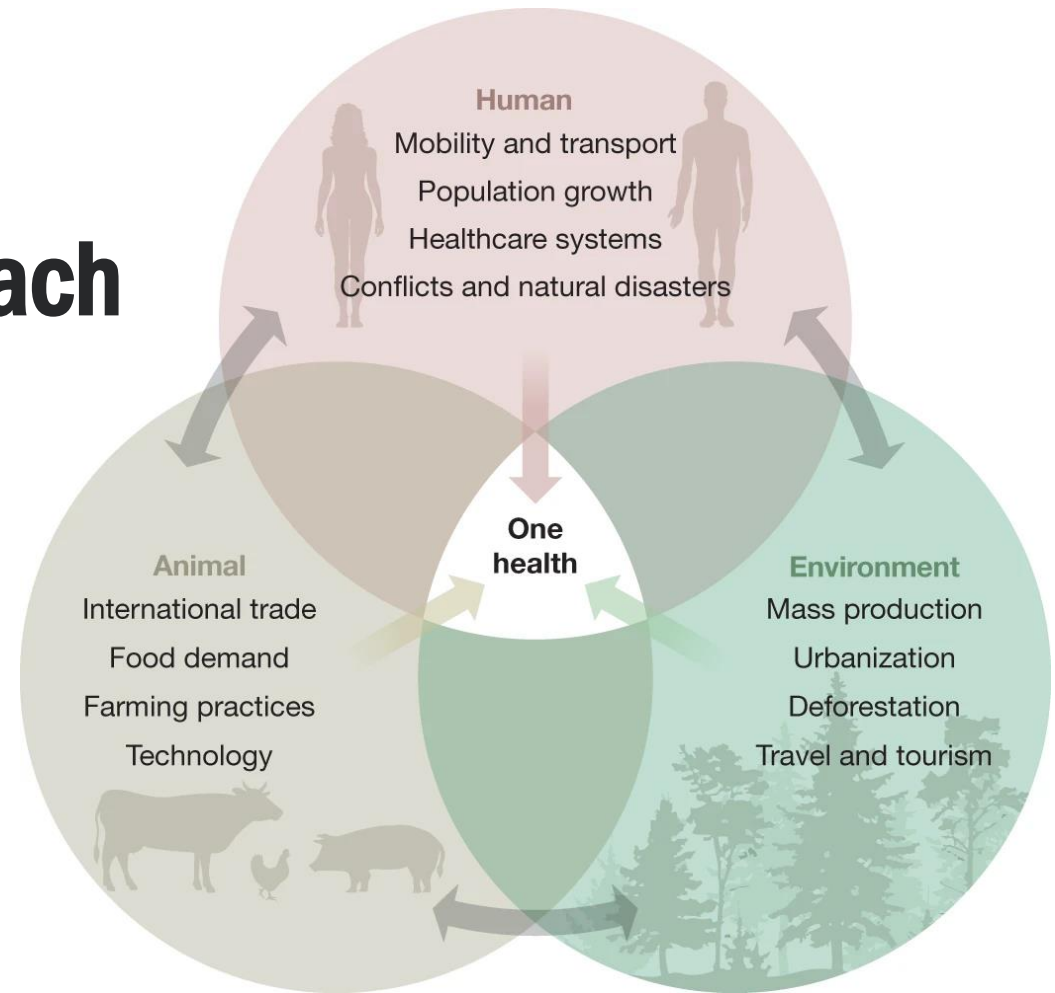
Pathogen dynamics in a changing world





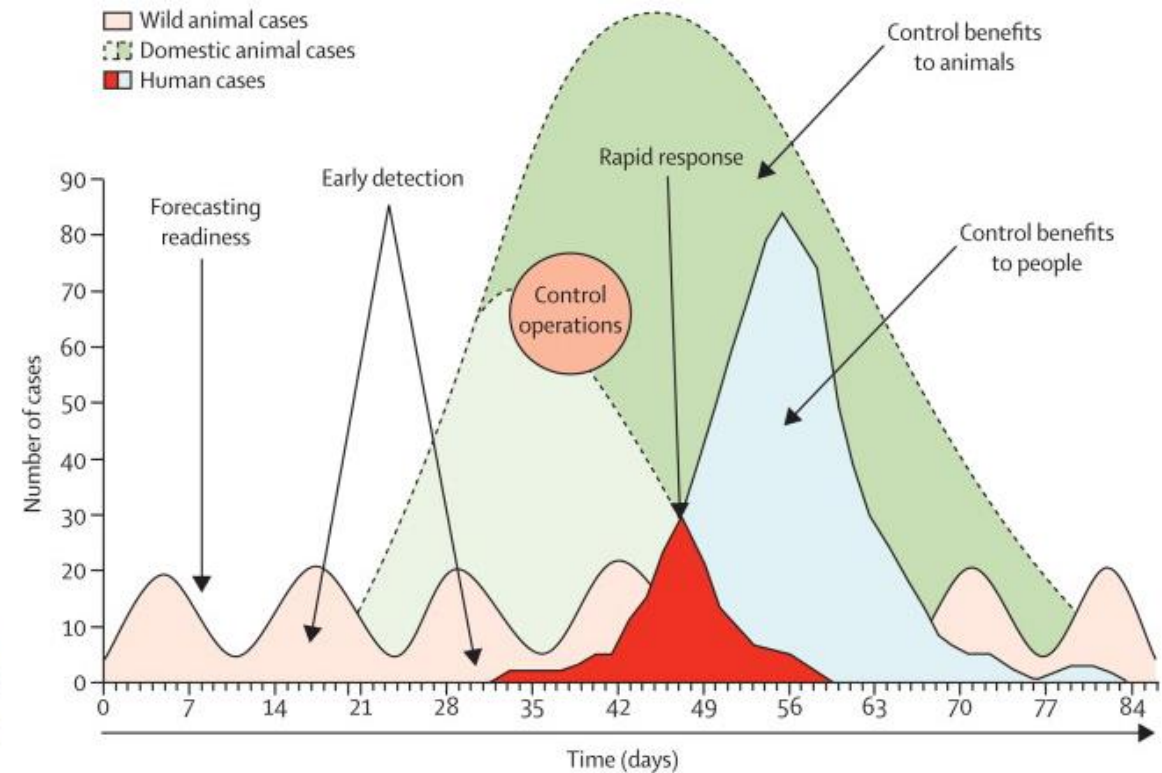
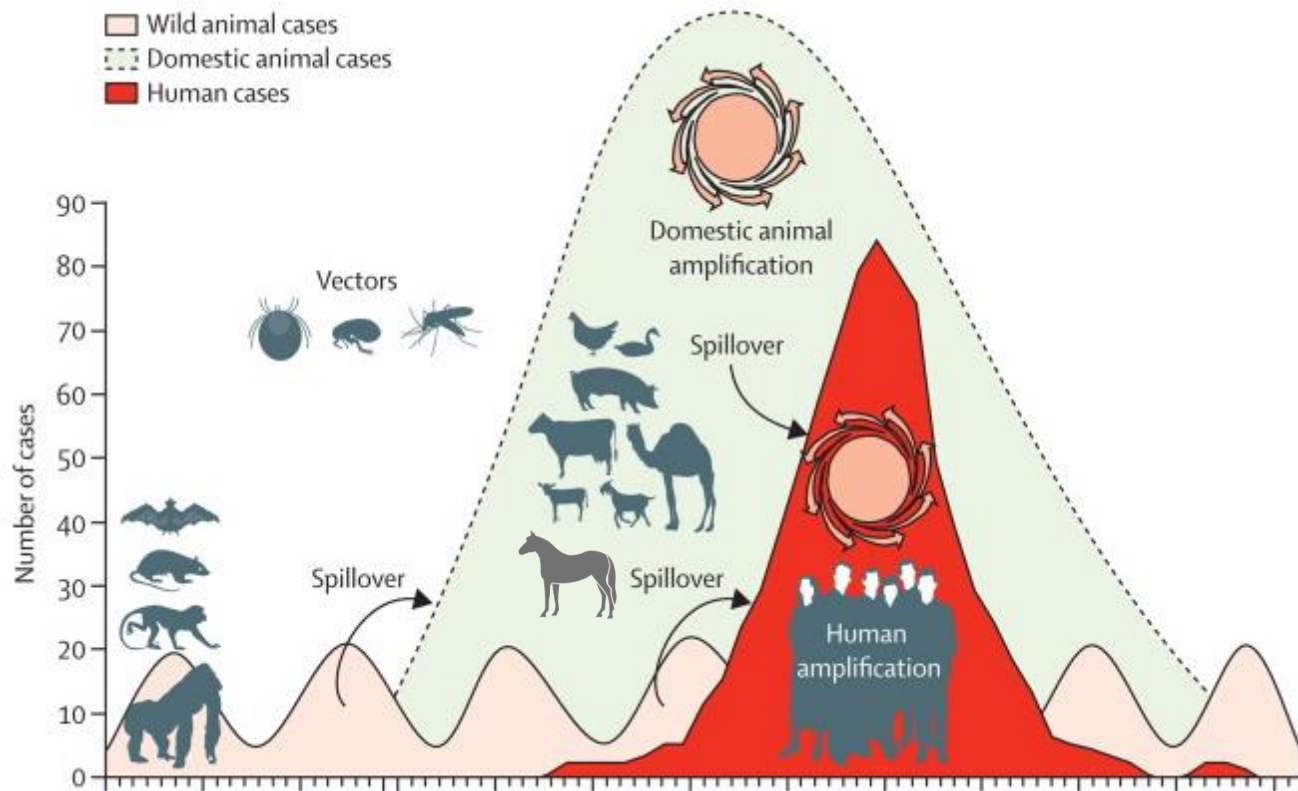
Adopting a **One Health** approach

“Human health and animal health are interdependent and bound to the health of the ecosystems in which they exist.”





Phases of pathogen emergence & surveillance



Overview of Vector-Borne Diseases (VBDs)

- **Importance:** VBDs significantly impact both human and animal health globally
- **Climate Change Impact:** Alters vector density, activity periods, and geographical distribution, increasing the risk of VBD spread
- The transmission potential of these diseases is **also influenced by a range of factors**, including socio-economics, health-care capacity and ecology.

Reporting of VBDs in 2023-2024 -Key Statistics:

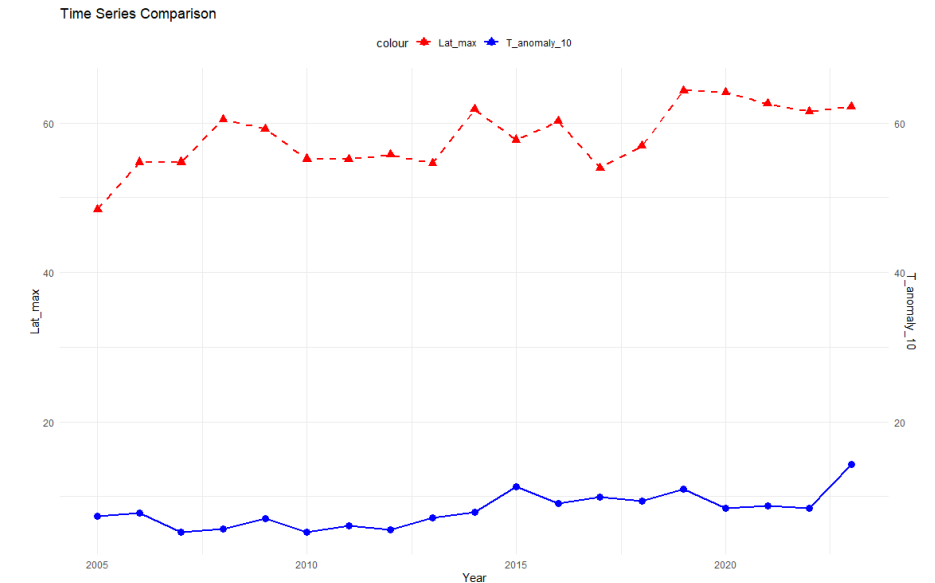
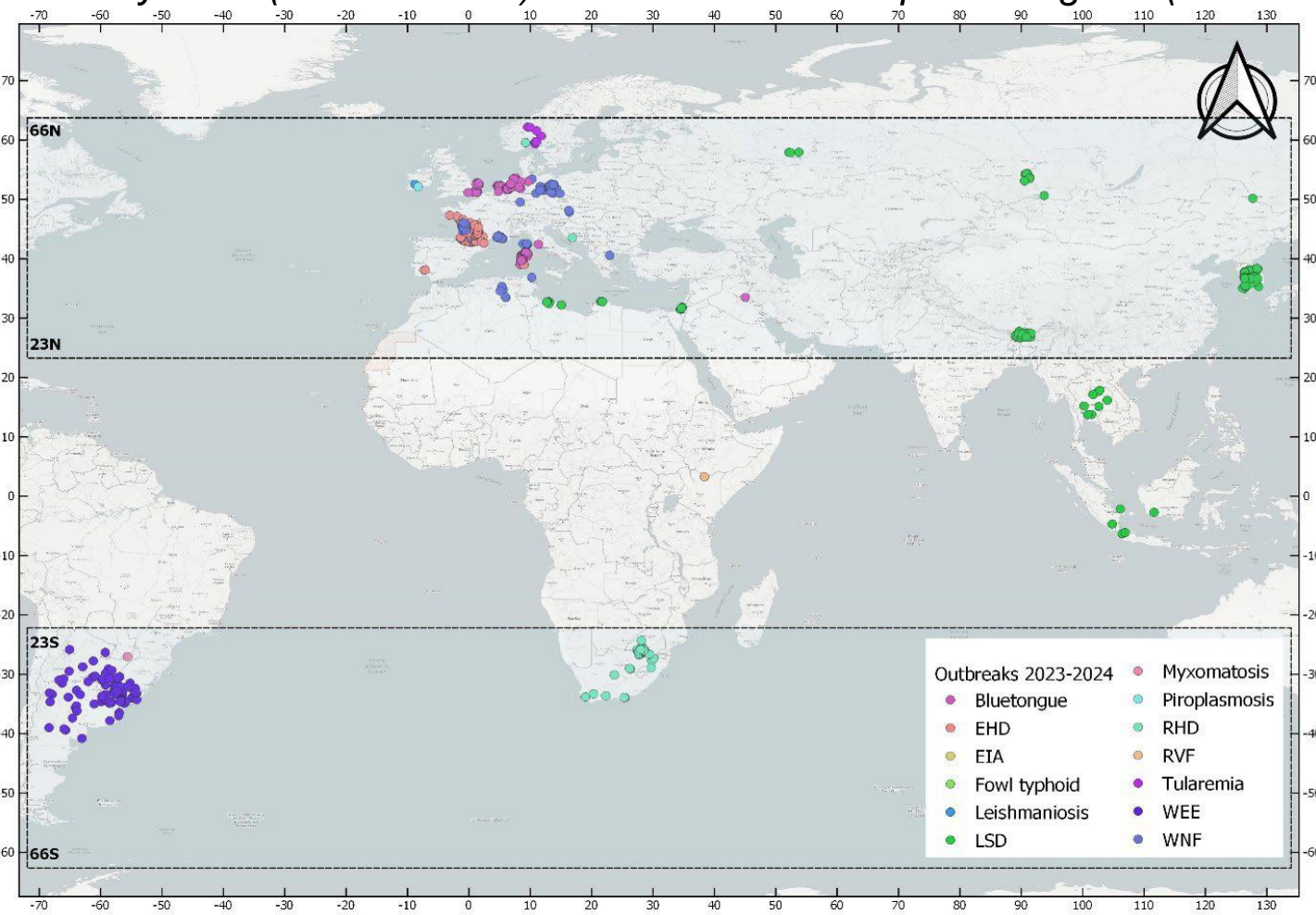
- **Most frequently reported:** West Nile Fever (12 notifications), Bluetongue (8), Lumpy Skin Disease (7), and Equine Infectious Anaemia (6).
- **Geographical Spread:** 28 countries reported 2,422 outbreaks, primarily in the Americas and Europe. Most of the outbreaks were concentrated in the Americas, with a very large event of **Western equine encephalomyelitis** (Western) (1,461 outbreaks), followed by Europe with 697 outbreaks, in this case reported for eight different VBDs.
- **Lumpy skin disease** was the most frequently reported VBD in Asia, with 144 outbreaks.



VBDs and Climate Change- Significant Observations:

- 99% of VBD outbreaks reported were in temperate regions.
- Increasing trend in the maximum latitude of VBD outbreaks, correlated with rising global temperatures.

Spatial distribution of outbreaks reported to WOAAH for ten VBDs through immediate notifications and follow-up reports during 2023 and early 2024 (as of 8 March). The limits of the temperate regions (23.5° and 66.5° N/S of the Equator) are shown on the map:

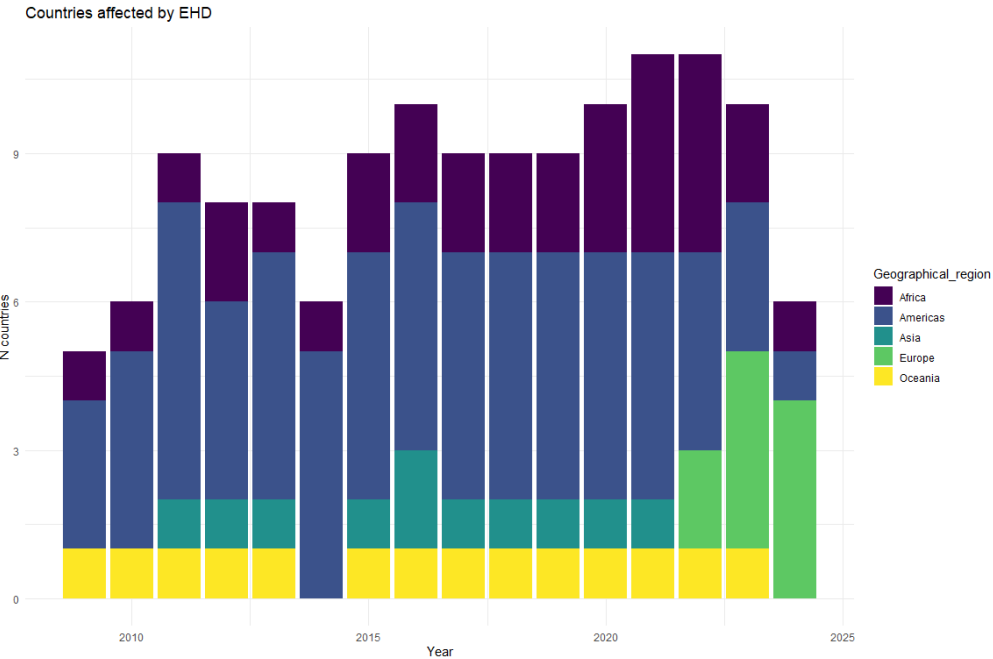
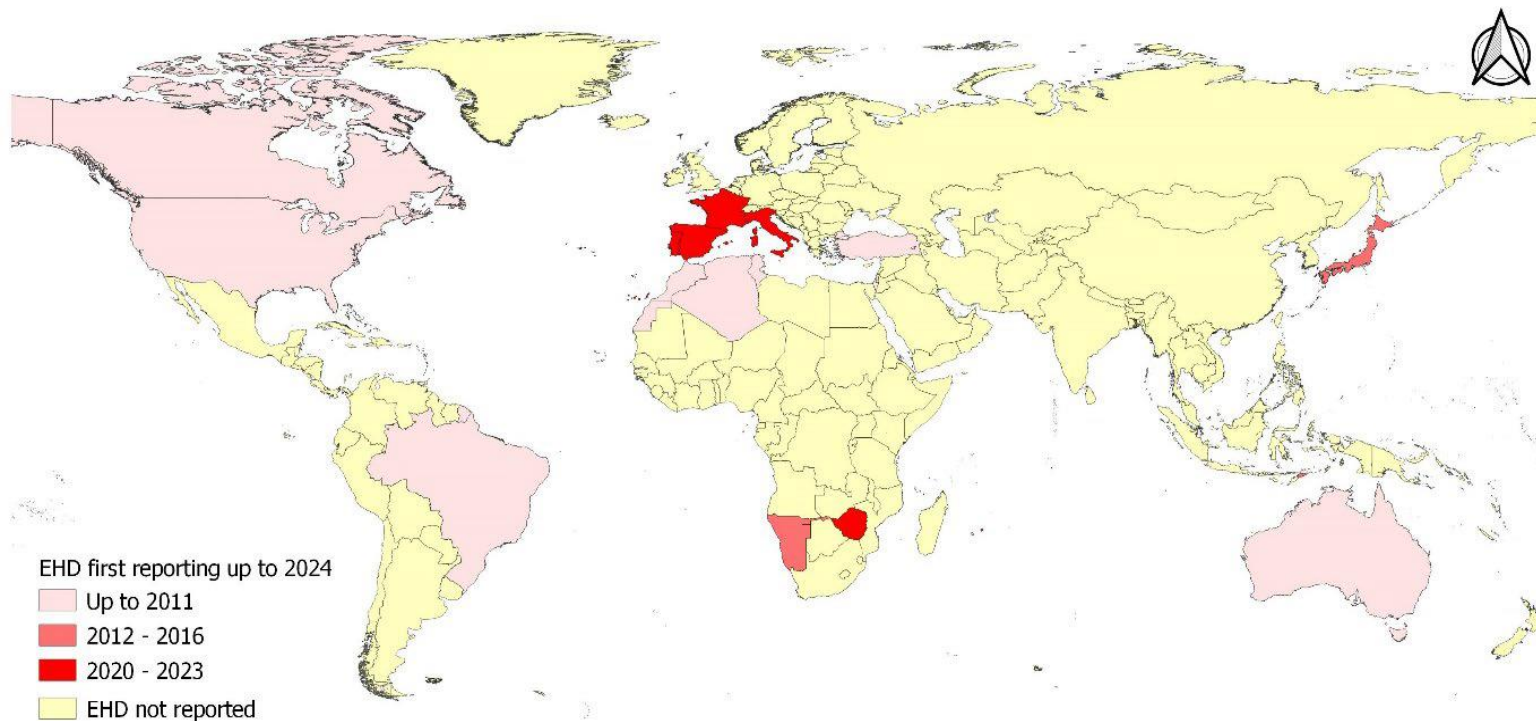


Time series comparison of the maximum annual latitude at which outbreaks were reported and the global annual anomaly temperature detection for the period 2005–2023.

Case Study: Epizootic Haemorrhagic Disease (EHD)

- **Spread:** EHD has expanded from North America to new areas, including Europe since 2022. At a global level, 23 countries have reported the presence of the disease since 2009.
- **Recent Developments:** 252 new outbreaks reported in Europe across four countries since 2022.

Global spread of EHD up to 2024. The map categorises countries based on the year of first reporting of the disease:



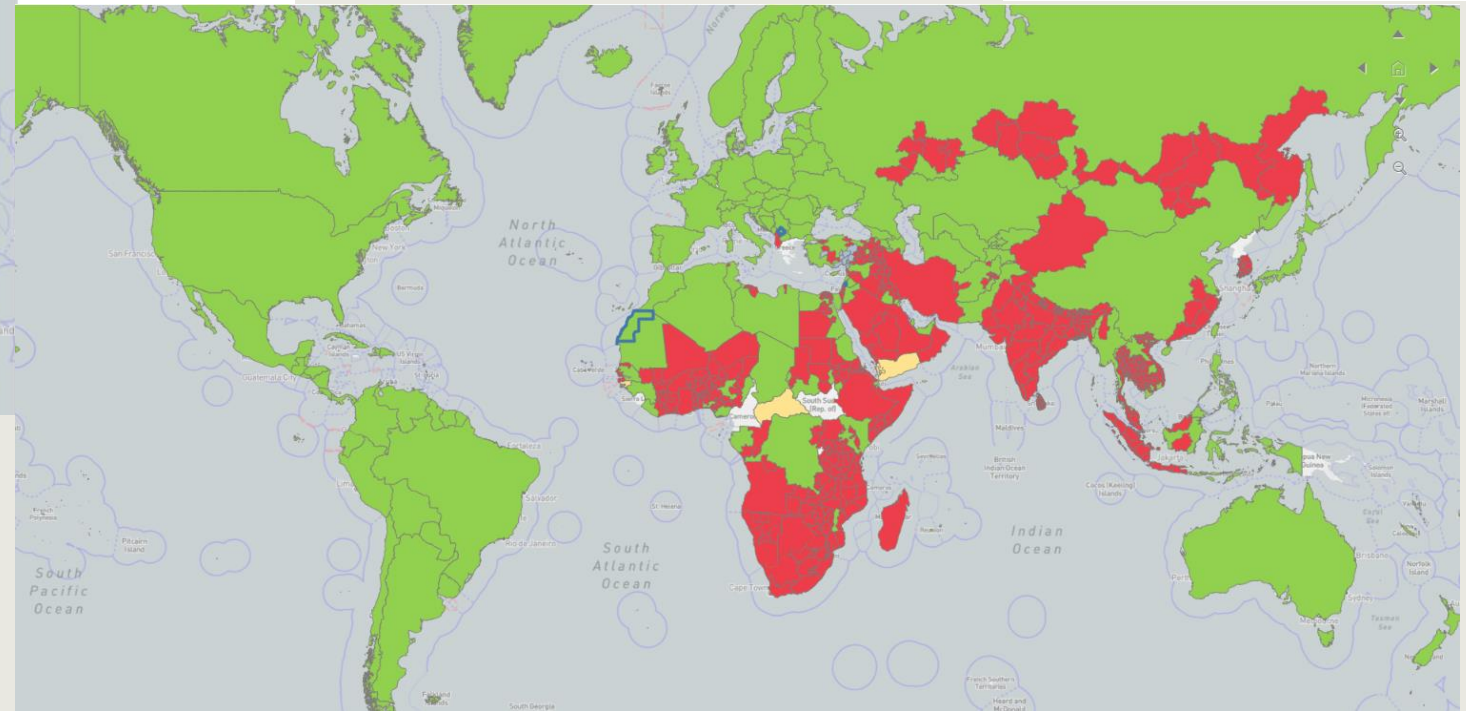
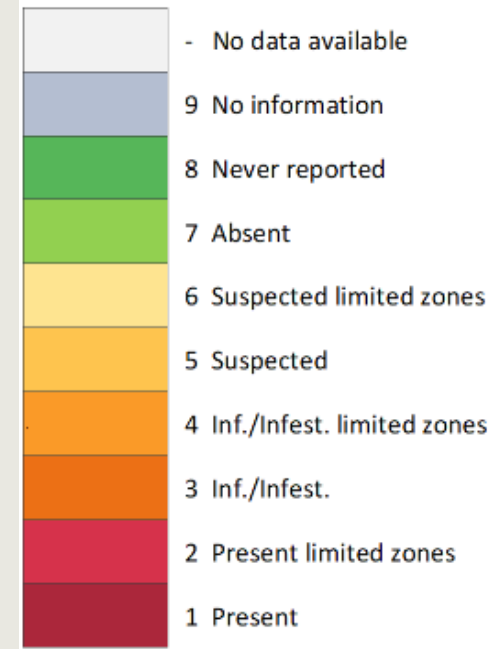
Evolution of countries reporting the presence of EHD during the period 2009–2024 (as of 8 March). Data for the different geographical regions are shown in the bar chart



DISEASE DISTRIBUTION: Lumpy Skin Disease- Data from [WAHIS](#) (2019-2024)

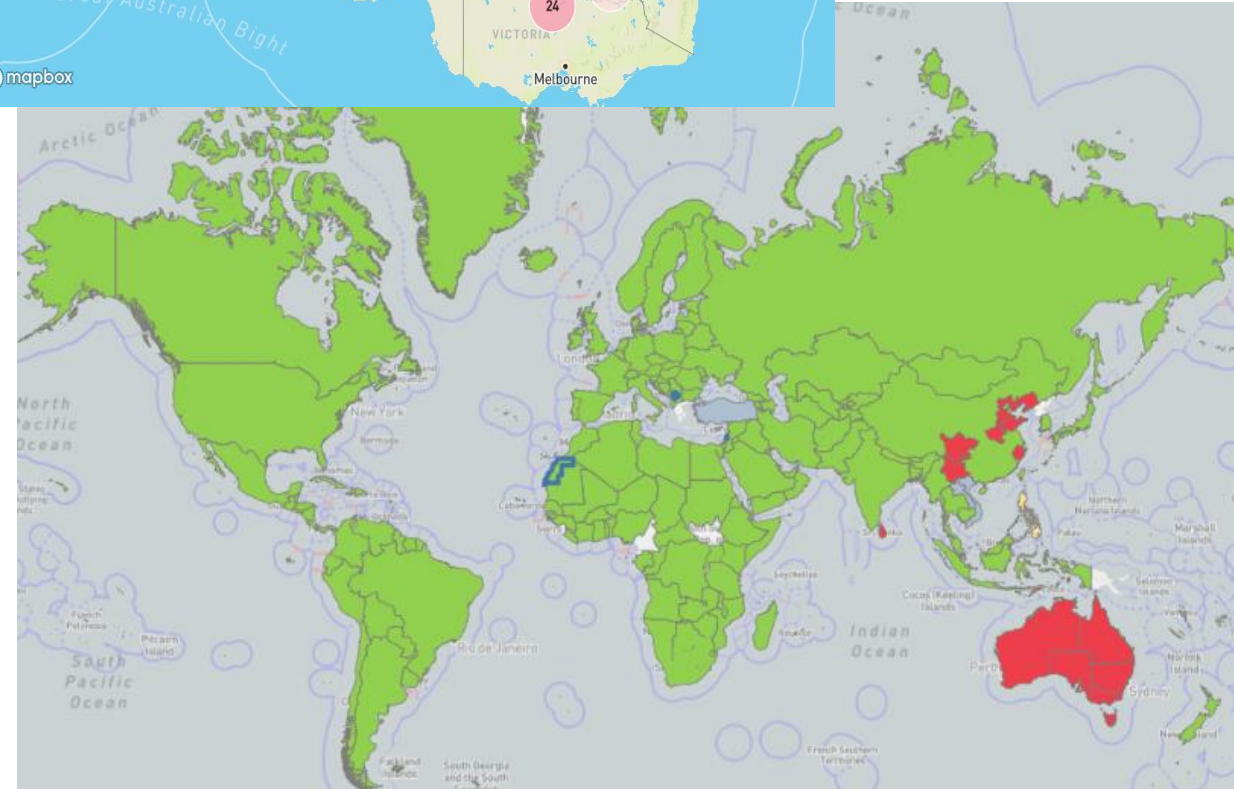


-  Domestic
-  Wild
-  Mixed
-  Resolved
-  Continuing
-  Cluster



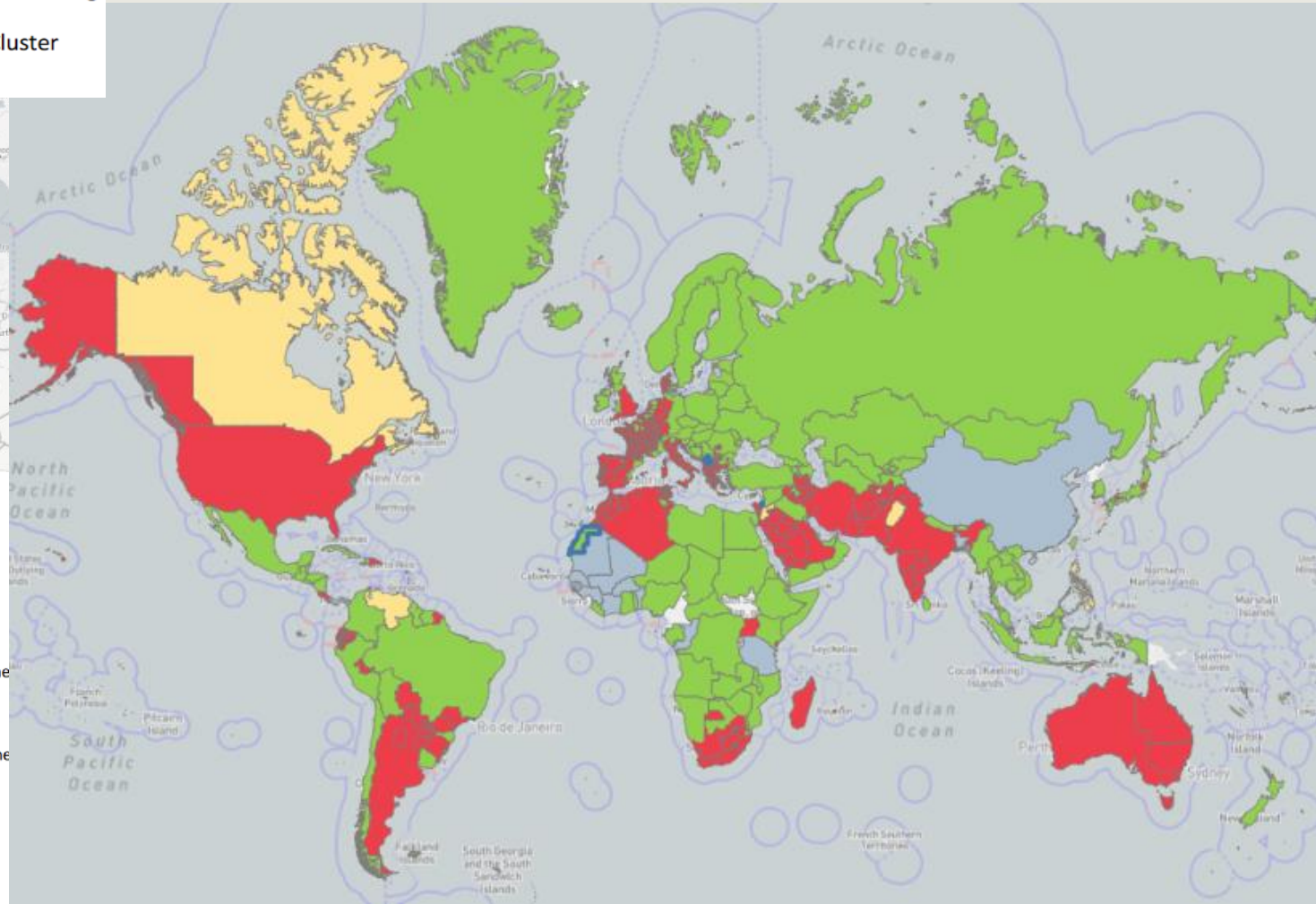
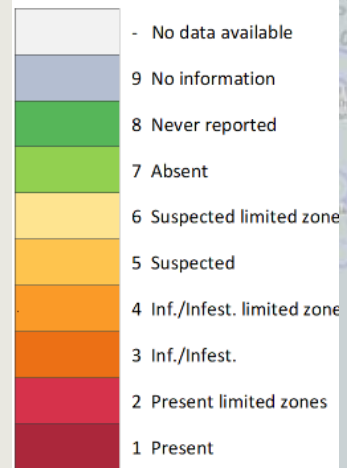
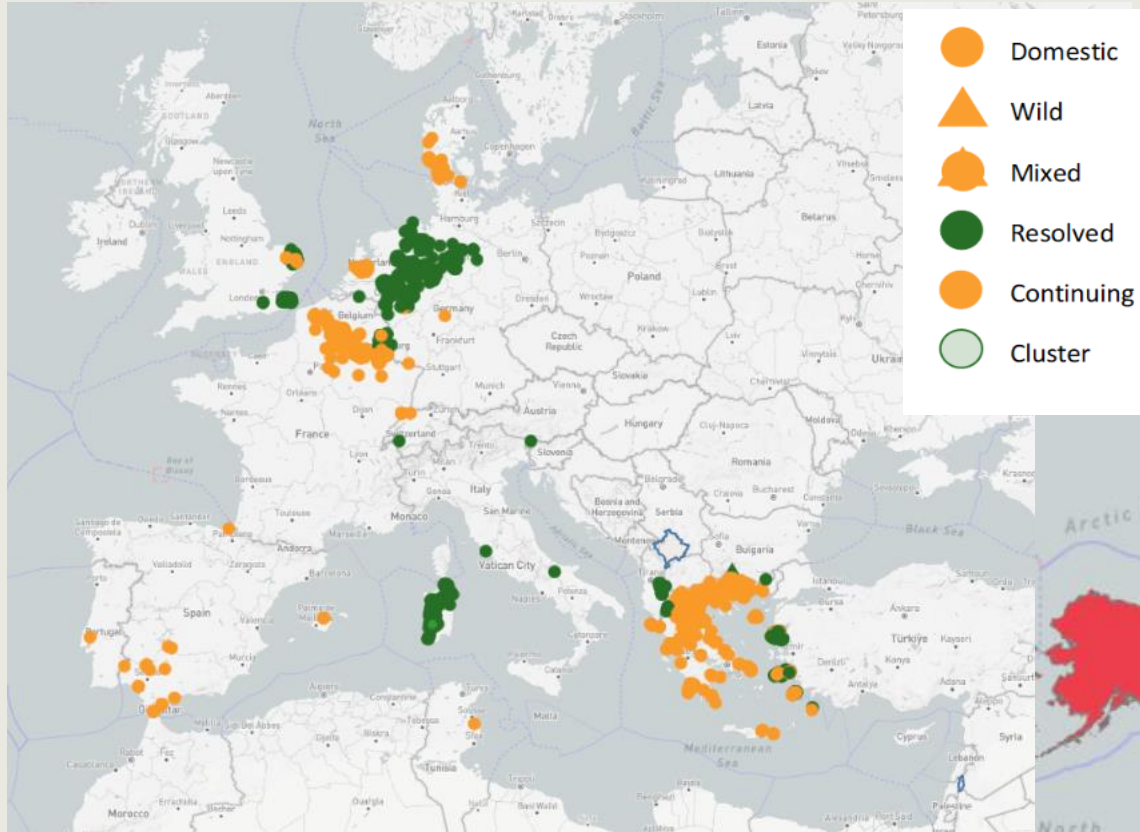


- Outbreaks of Japanese encephalitis have been reported in piggeries in Queensland, New South Wales, Victoria and South Australia.
- Climate conditions of above median rainfall and warmer minimum temperature may have been a factor in the event.
- Spillover events into Humans and a case in an Alpaca
- Infection with JE virus in horses were not detected
- Trade restrictions for the movement of horses according to WOHAI provisions triggered the revision of the JE Code Chapter





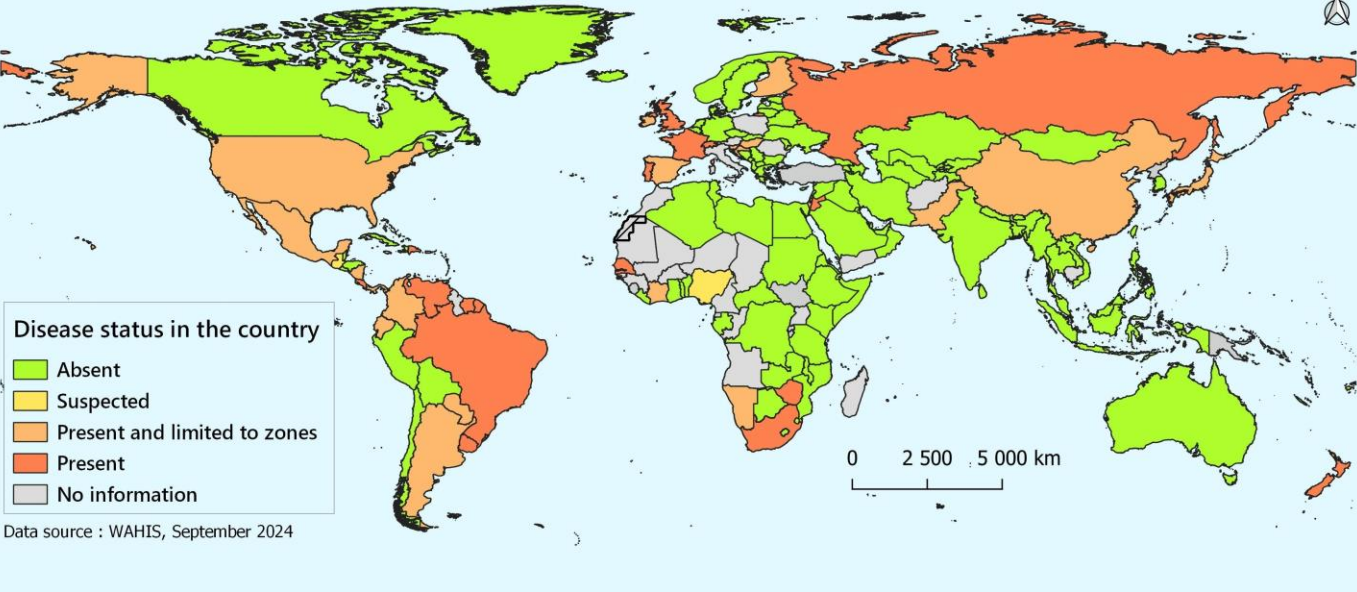
DISEASE DISTRIBUTION: Bluetongue- Data from [WAHIS](#) (2019-2024)



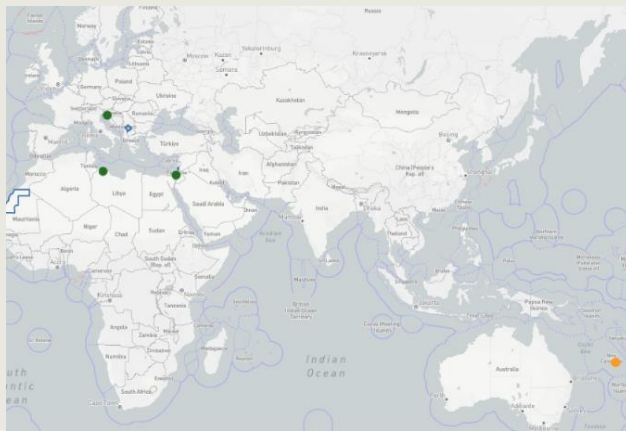
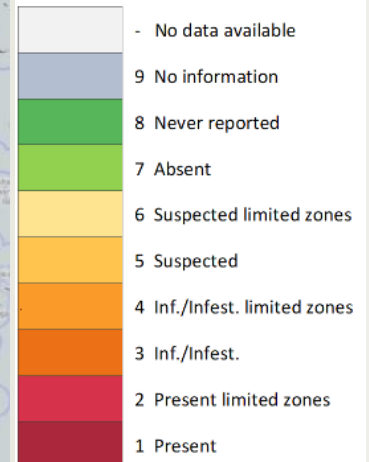
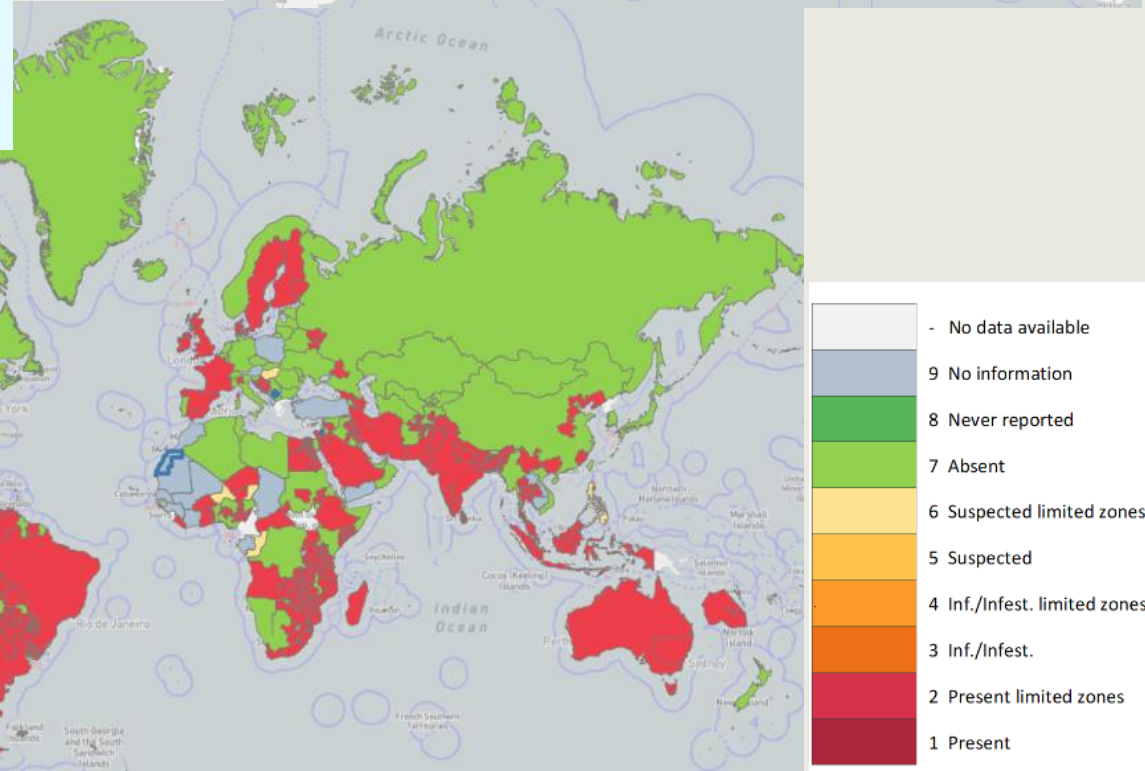
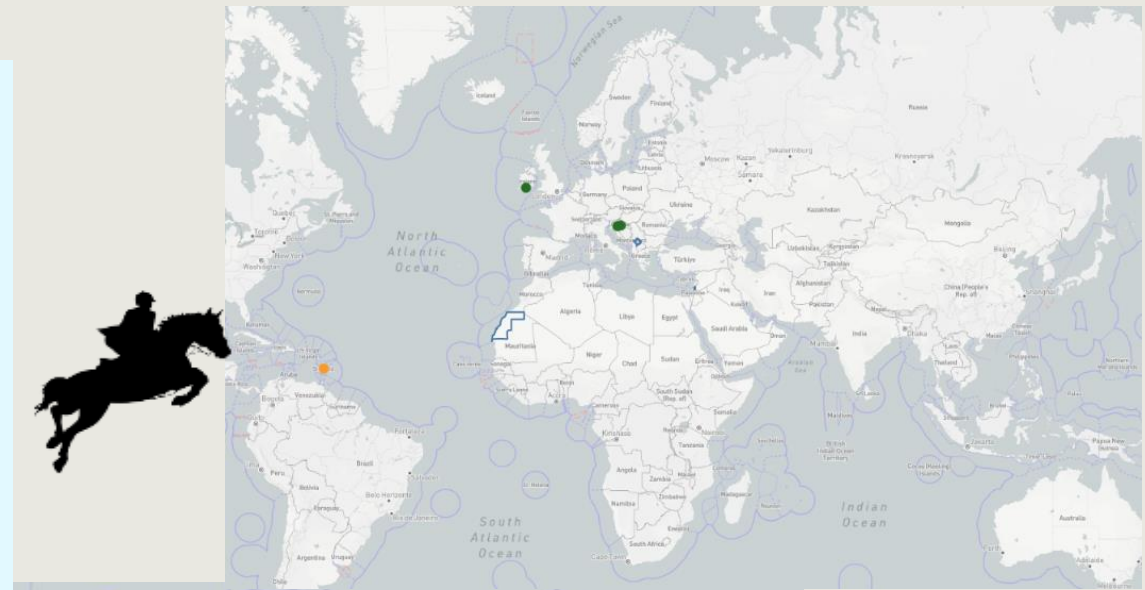


DISEASE DISTRIBUTION: Equine Piroplasmosis & Bovine Babesiosis– Data from [WAHIS](#)

Global distribution of equine piroplasmosis (2019-2023)

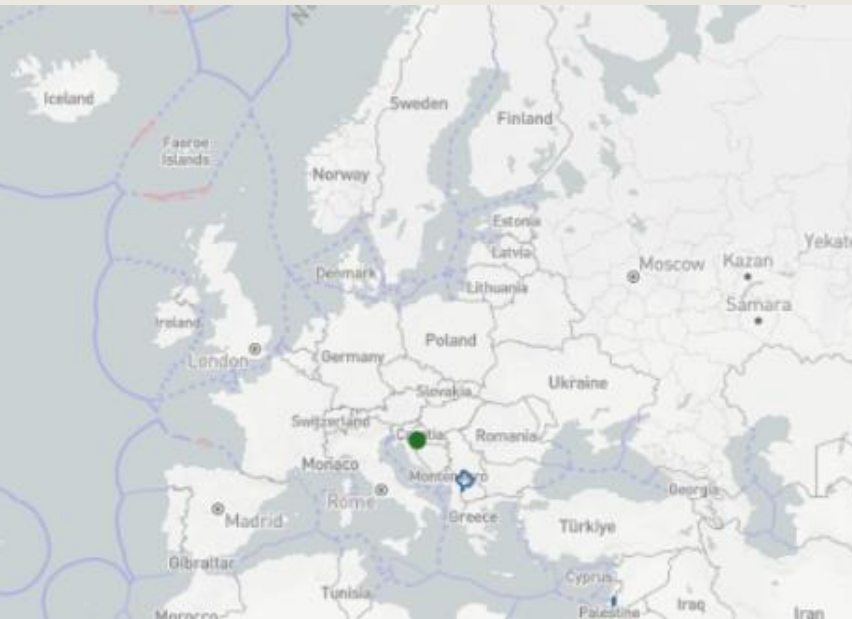
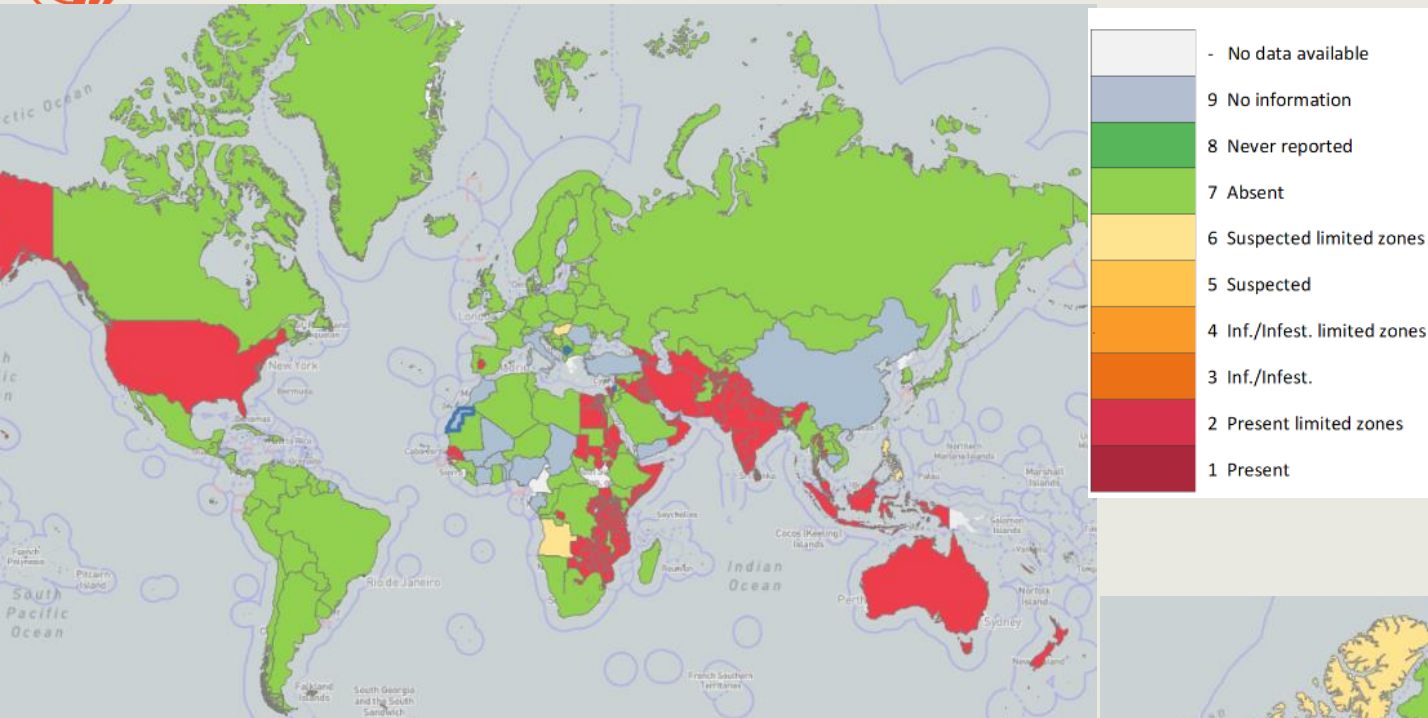


Data source : WAHIS, September 2024

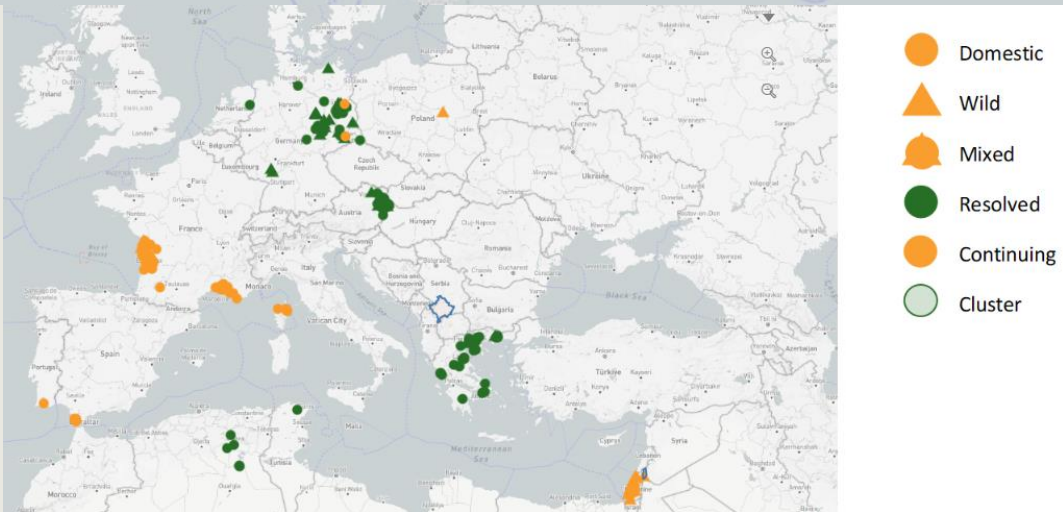
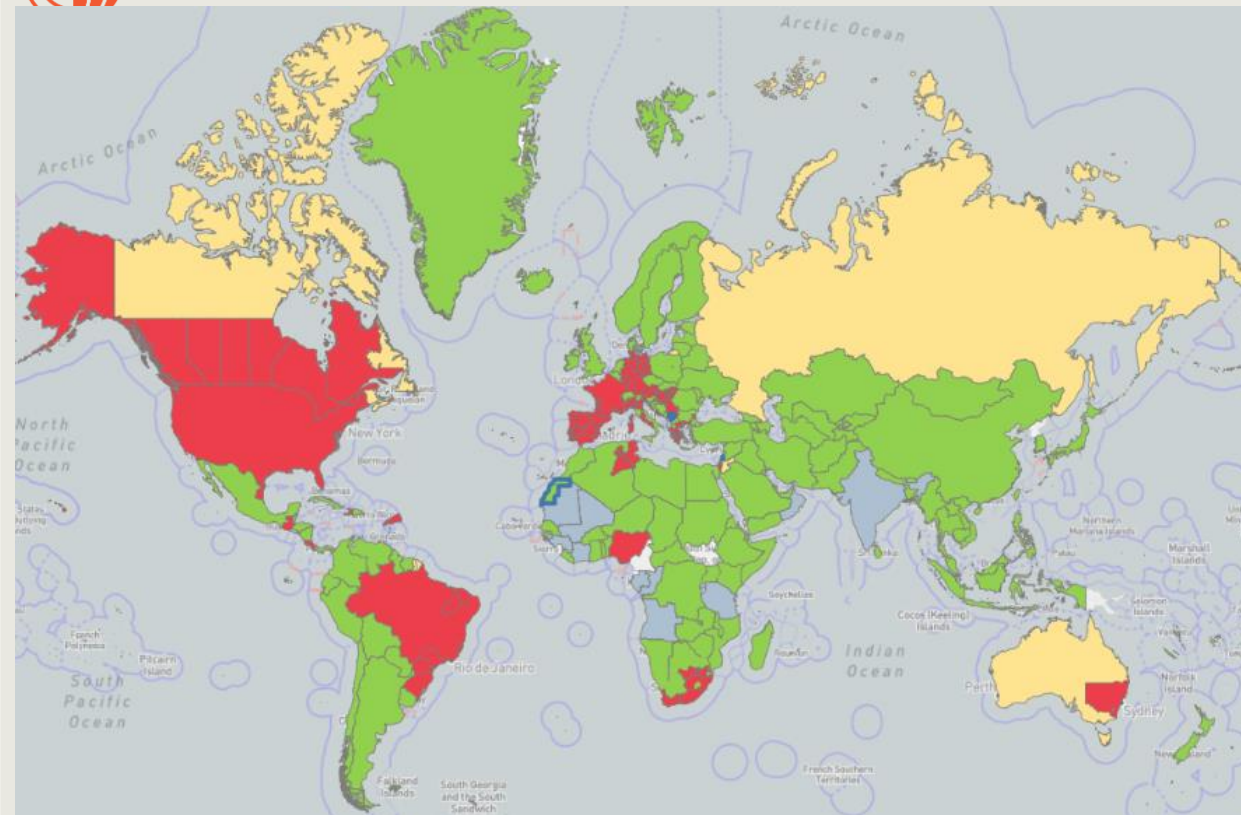




DISEASE DISTRIBUTION: Theileriosis (left above) & Bovine Anaplasmosis (below) – Data from [WAHIS](#)

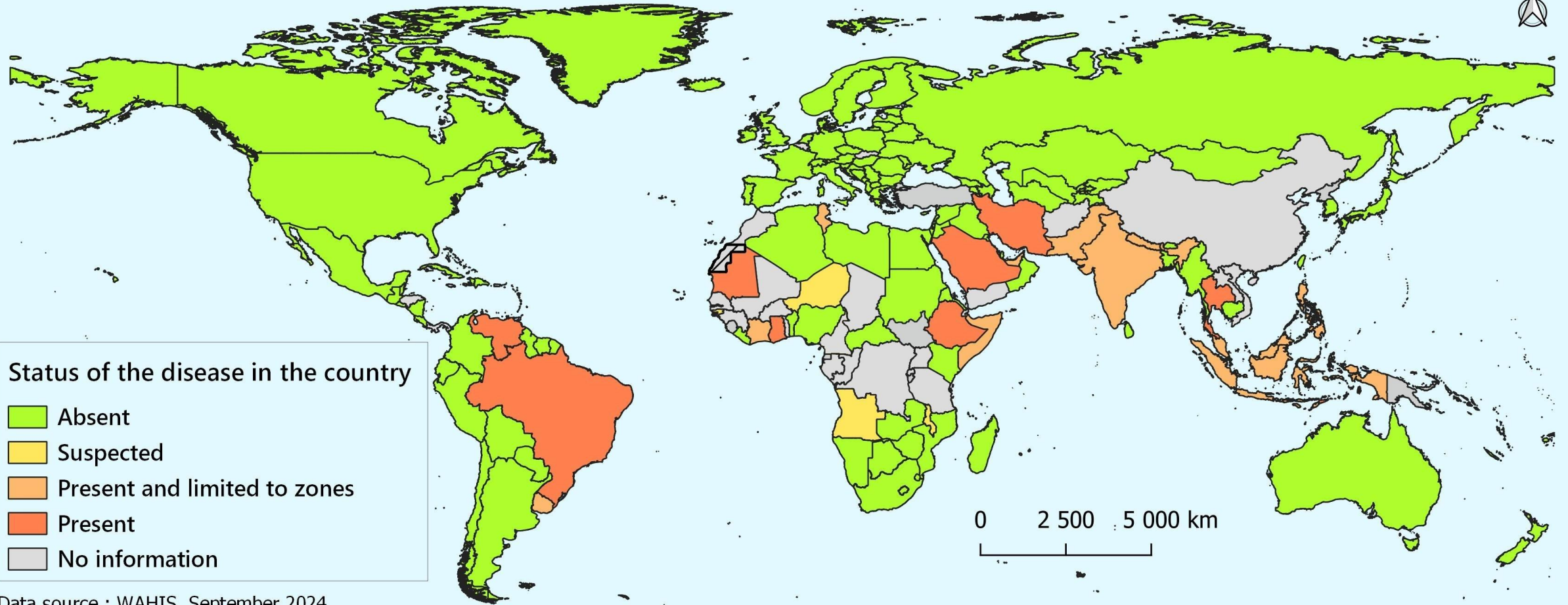


DISEASE DISTRIBUTION: West Nile Fever (left) & Western Equine Encephalomyelitis (right) - [WAHIS](#)





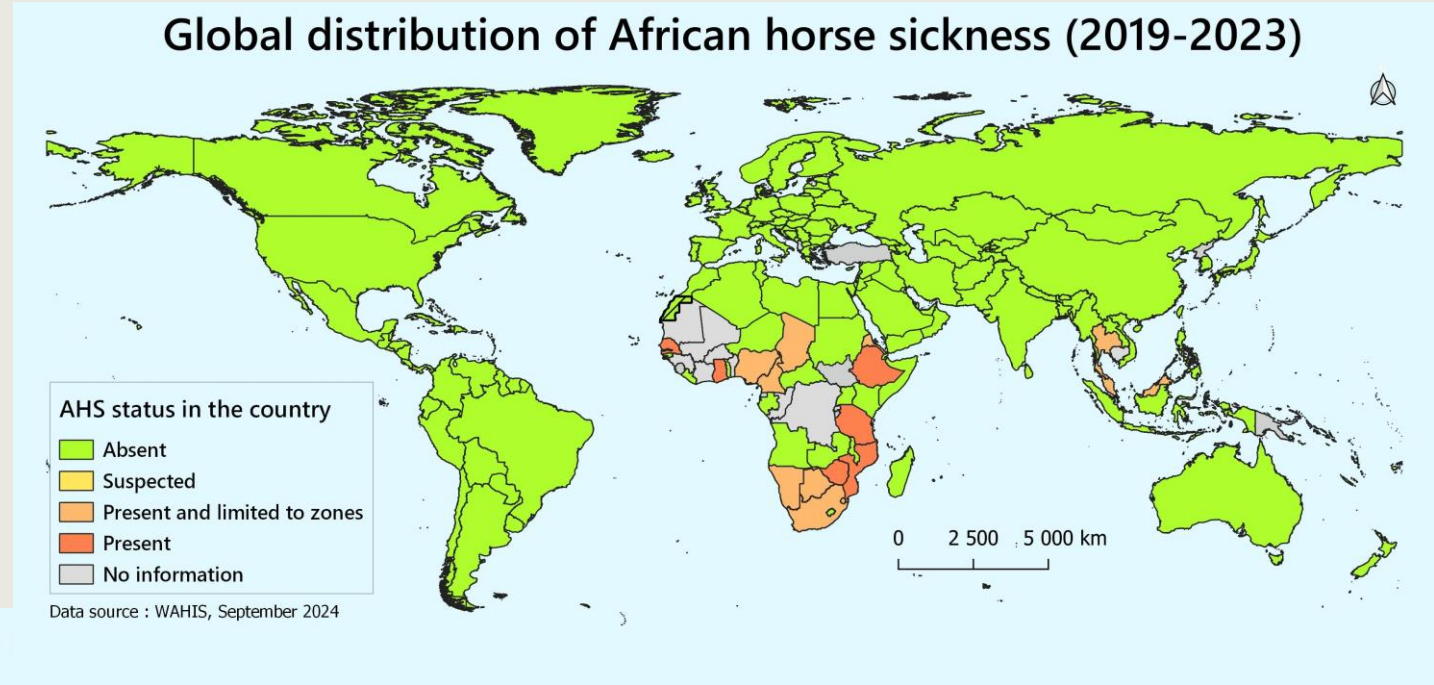
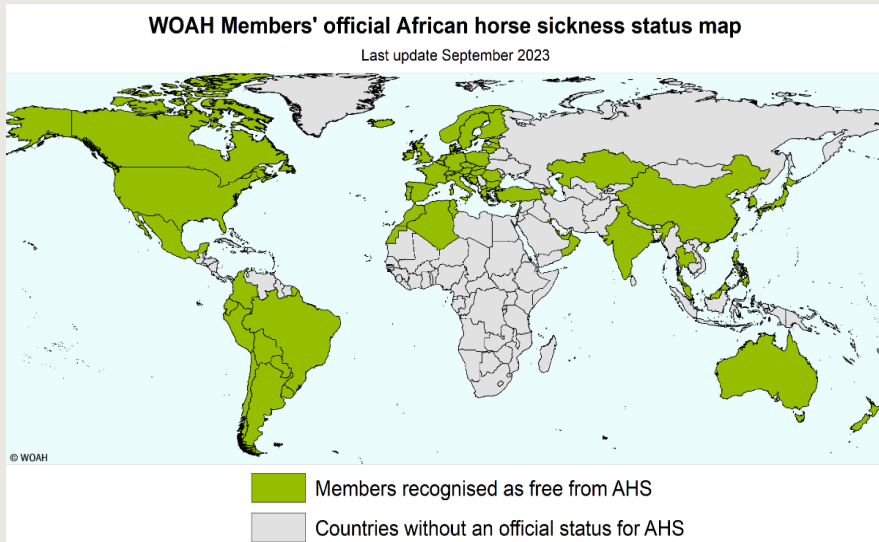
Global distribution of Surra (*Trypanosoma evansi*) (2019-2023)



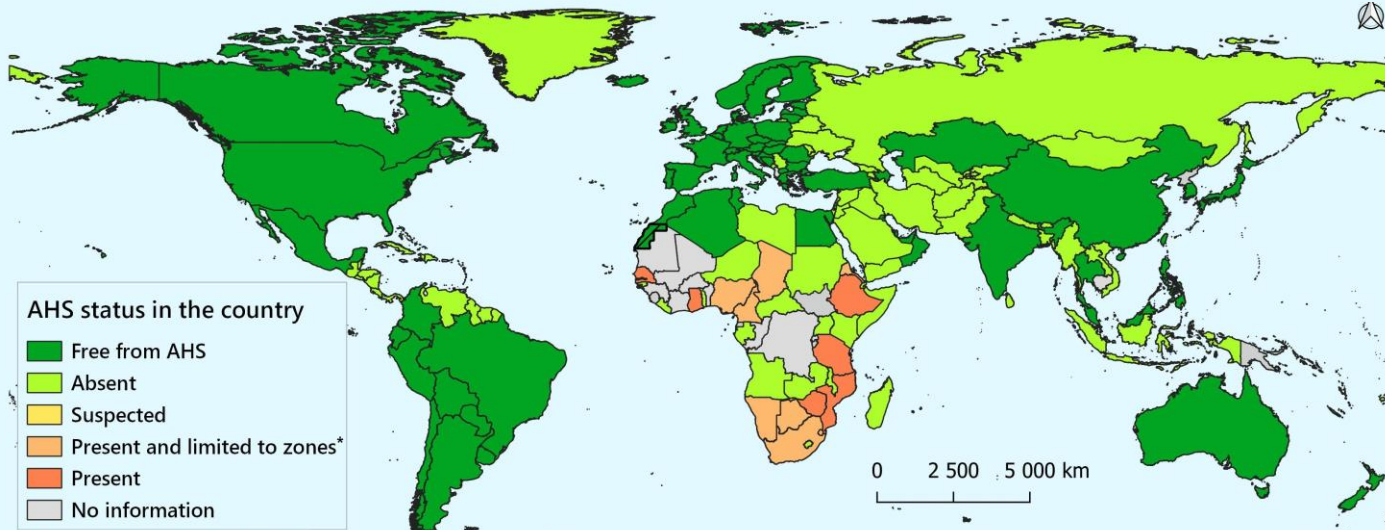
Data source : WAHIS, September 2024



DISEASE DISTRIBUTION - Data from the World Animal Health Information System (WAHIS)



Global distribution of African horse sickness



*AHS notified in Malaysia (Trengganu) in August 2020
Data source : WAHIS, September 2024



Key Takeaways:

Climate change is a major driver in the spread of VBDs

- Expanded Geographical Range
- Increased Vector Activity
- Unpredictable Outbreaks
- Correlation with Global Warming:

Recommendations

- Heightened Disease Surveillance
- Need for Global Collaboration
- Health and Economic Impacts

Continuous monitoring and adaptive strategies are crucial for effective VBD management.





Thank you

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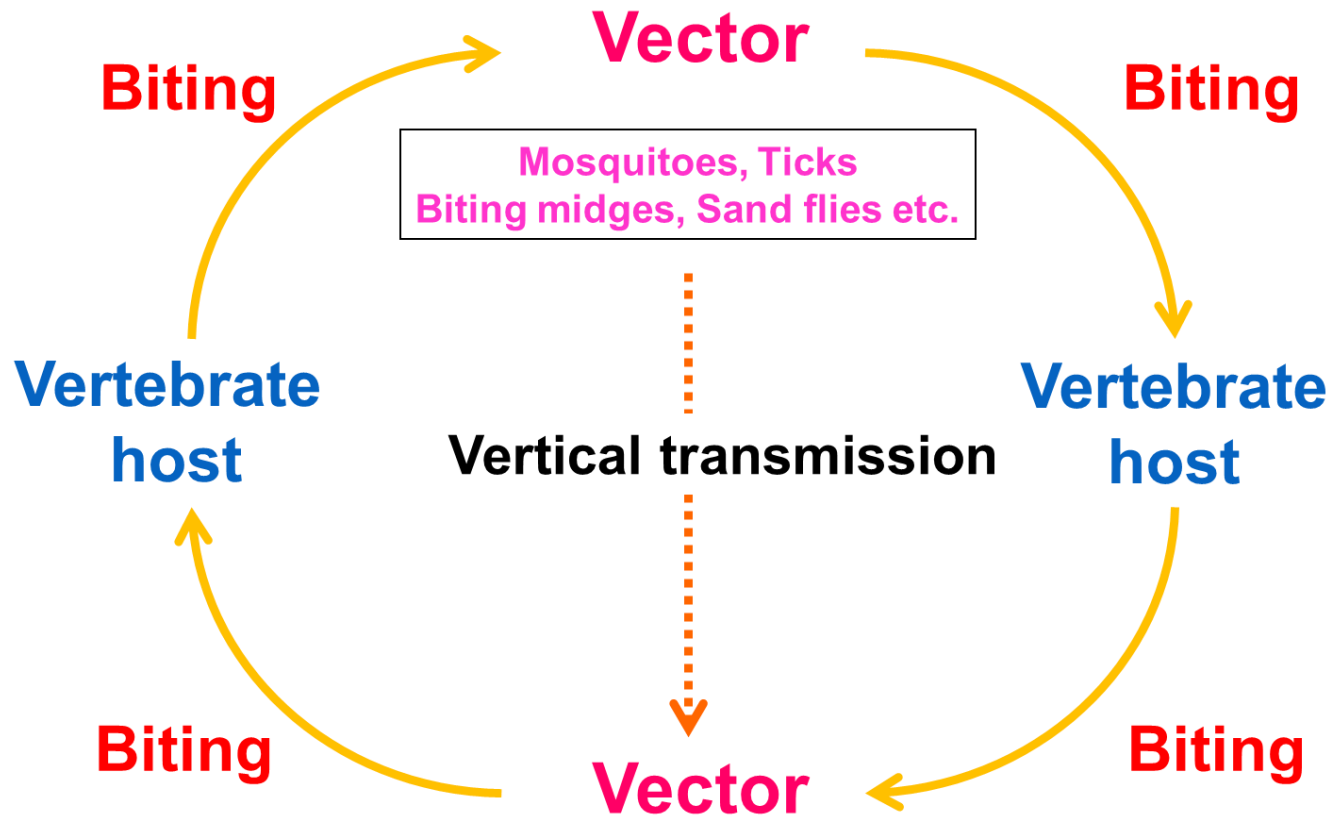
Arbovirus infections of livestock animals in Asia: what we know and don't know

Tohru Yanase

Kagoshima Research Station,
National Institute of Animal Health, NARO,
Japan

Arbovirus (Arthropod-borne virus)

- Arboviruses are transmitted by arthropod vectors via blood sucking process.
- Arboviruses replicate in both arthropod vectors and vertebrate hosts.
- Arboviruses include many taxonomically different virus groups.



WOAH-listed arbovirus infections in Asia

Infection	Affected animal	Zoonosis	Endemic in Asia	Vector
Bluetongue	ruminants	No	Yes	<i>Culicoides</i>
Epizootic hemorrhagic disease	ruminants	No	Yes	<i>Culicoides</i>
African horse sickness	horse	No	No	<i>Culicoides</i>
Japanese encephalitis	pig, horse	Yes	Yes	mosquitos
Nairobi sheep disease	sheep	No	Yes	ticks
Crimean–Congo hemorrhagic fever	ruminants*	Yes	Yes	ticks
African swine fever	pig	No	Yes	ticks**

* Subclinical in most cases

** Highly contiguous between animals without ticks

- We don't know the past and present status of the listed arbovirus infections in detail.

Due to

Lack of accurate information of affected animals, epidemiology, etiology and so on.....

Bluetongue (BT):

- Infection with BT virus and affecting ruminants
- High mortality rate in sheep of up to 70%
- Reported sporadic outbreaks in Asia

Epizootic hemorrhagic disease (EHD):

- Infection with EHD virus
- Severe clinical symptoms in white tailed deer and cattle
- Large outbreaks of Ibaraki disease with deglutition disorder in Japan

- BT and EHD viruses have been circulating in Asia.
- Multiple serotypes of these viruses are prevalent.

We don't know

How many animals are affected by both infections (no good statistical data).

Which serotypes and/or strain has high virulent to domestic animals.

Which vector species principally transmit the viruses in each region.

Not listed, but important arbovirus infections in Asia

Infection	Affected animal	Manifestations	Vector
Akabane disease	ruminants	abortion, premature birth, congenital malformations	<i>Culicoides</i>
Aino virus infection	ruminants	abortion, premature birth, congenital malformations	<i>Culicoides</i>
Chuzan disease	cattle	congenital malformations	<i>Culicoides</i>
Bovine ephemeral fever	cattle, water buffalo	cessation of lactation in dairy cattle, loss of condition in beef cattle	mosquito or <i>Culicoides</i>
Getah	horse, pig	fever, edema, rash (horse) reproductive disorders (pig)	mosquito

- The causative viruses have been isolated/detected in Asian countries/regions.
- These arbovirus infections have impacted livestock industry for many years.

The current status of these arbovirus infections are not shared in regional and global levels.

Akabane disease

Etiological agent:

Akabane virus

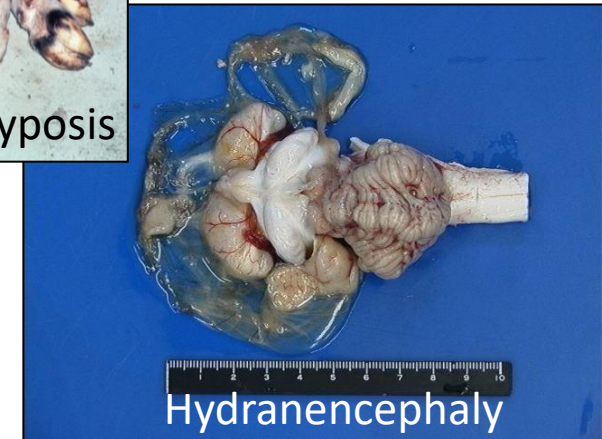
Genus: *Orthobunyavirus*

Symptoms:

- Abortion, premature birth, still birth, congenital abnormalities (in utero infection)
- Encephalomyelitis (postnatal infection)

Affected hosts:

Cattle, water buffalo, sheep, goat



- In the past large outbreak, 42,000 cattle were affected in Japan, indicating Akabane disease has a high potentiality to impact livestock industry.
- Akabane virus is widely distributed in Asia, but there is a few report of affected animals from Asian countries and regions, except Japan.

Arboviruses potentially affecting livestock industry in Asia

Genus	Virus	Affected animal	Manifestations (suspected)	Vector
<i>Orthobunyavirus</i>	Peaton virus	cattle, sheep	abortion, congenital malformations	<i>Culicoides</i>
	Shamonda virus	cattle, goat	abortion, congenital malformations	<i>Culicoides</i>
	Sathuperi virus	cattle	congenital malformations	<i>Culicoides</i>
<i>Orbivirus</i>	D'Aguilar virus	cattle	congenital malformations	<i>Culicoides</i>


Orbiviruses recently identified in East Asia

Virus	Location of first detection in East Asia	Year of isolation	Vector
EHDV serotype 5	Japan	2016	<i>Culicoides</i>
EHDV serotype 6	Japan	2014	
EHDV serotype '10'	Japan	1998 (2017)	
EHDV YNDH/V079/2018	China	2018	
BTV serotype 29	China	2014	
BTV-X/XJ1407	China	2014	
Tibet orbivirus serotype 1	China	2009	
Tibet orbivirus serotype 2	China	2007	
Tibet orbivirus serotype 3	Japan	2009	
Tibet orbivirus serotype 4	Japan	2010	
Tibet orbivirus serotype 5	China	2019	
Tibet orbivirus serotype 6	China	2019	
Bunyip Creek virus	Japan	2008	
Marrakai virus	Japan	1997 (2024)	
Yunnan orbivirus	China	1999	mosquito
Guangxi orbivirus	China	2015	
Yonaguni orbivirus	Japan	2014	

an outbreak of EHD in Japan in 2015

suspected association with neurological signs in cattle and sheep in Peru

EHD: epizootic hemorrhagic disease
BT: bluetongue

 Initially isolated in East Asia

Potential impact of neglected and emerging arboviruses

- It is difficult to assess the impact of neglected and emerging arboviruses

Lack of

Characterization

Diagnostic and detection systems

Epidemiological information



We should be concerned with
neglected and emerging
arboviruses more!

Mutation

- RNA viruses are characterized by high mutation rates.
- Mutations may change pathogenicity, antigenicity, host range and competency of vectors.

A specific genotype of Akabane virus caused bovine encephalomyelitis by postnatal infection

Porcine reproductive failures associated with Akabane virus were recently reported

Reassortment

- Related segmented viruses exchange genome segments in co-infected cells. The process is called “reassortment”.
- Reassortment may change pathogenicity, antigenicity, host range and competency of vectors, drastically.

Orthobunyavirus

Genome:

three segmented, negative-sense,
single-stranded RNA

Akabane virus, Aino virus,
Schmallenberg virus

Orbivirus

Genome:

ten segmented, double-stranded RNA

Bluetongue virus, Chuzan virus,
Epizootic hemorrhagic disease virus

Zoonotic arboviruses

Virus	Manifestations in human	Vector
Batai virus	mild flu-like illness	mosquito
Banna virus	encephalitis	mosquito
Japanese encephalitis virus	encephalitis	mosquito
SFTSV	severe fever with thrombocytopenia syndrome	tick
Crimean–Congo hemorrhagic fever virus	hemorrhagic fever	tick
Kyasanur forest disease virus	hemorrhagic fever	tick
Tick-borne encephalitis virus	encephalitis	tick

- Several mosquito- and tick-borne viruses infect both human and livestock animals.
- Livestock animal may play a role of amplifier for zoonotic arboviruses in some cases.

“Contribution” of livestock animals in transmission cycle of several zoonotic arboviruses remains uncertain.

Global warming

- Global warming may affect distribution, migration, phenology and competence of vectors.



Expansion of distribution range of arboviruses
Changing epidemiology of arbovirus infections

Vector studies are not enough in Asian.

Little information

Principal vectors of each virus in each region

Biology of vector species

Assessment of vector competence by laboratory experiment

Essential for future advancement

- Considering the impact of non-listed arbovirus infections (such as Akabane disease and bovine ephemeral fever)
- Risk assessment of neglected and emerging arboviruses
- Collaboration between public health and veterinary regions
- Information sharing with neighboring countries
- Constructing functional reporting systems for arbovirus infections
- Human resources development for arbovirus and vector researches
- Supports from our governments and WOAHA

Thank you for your attention!!



Bluetongue virus

Update: Global and Regional
and new advances in virus
tracking

Dr Stacey Lynch

CSIRO Australian Centre for Disease
Preparedness . **WOAH Reference Laboratory for
Bluetounge virus**

WOAH Regional Workshop on VBDs in Asia and
the Pacific | September 2024
Australia's National Science Agency





ACDP provides the highest levels of biocontainment within a purpose built biosecurity infrastructure to help protect Australia's livestock and aquatic industries, as well as people, from emerging infectious disease threats.

WOAH Reference Laboratory for Bluetounge virus : global ecology (esp. regional) inc. molecular epidemiology, improved diagnostics and surveillance strategies to support industry and trade.

- **Dr Debbie Eagles, Director ACDP**
- **Dr Tristian Read, Veterinarian, ACDP**





Bluetongue virus



The Disease

- Arbovirus – *Culicoides* midges the primary vector
- A severe viral disease primarily affecting sheep, however infections also occur in cattle, goats and camelids
- WOAH-listed disease – Important implications for trade
- Australia claims freedom from clinical bluetongue disease, however bluetongue virus transmission is common in northern NSW, QLD, NT and WA

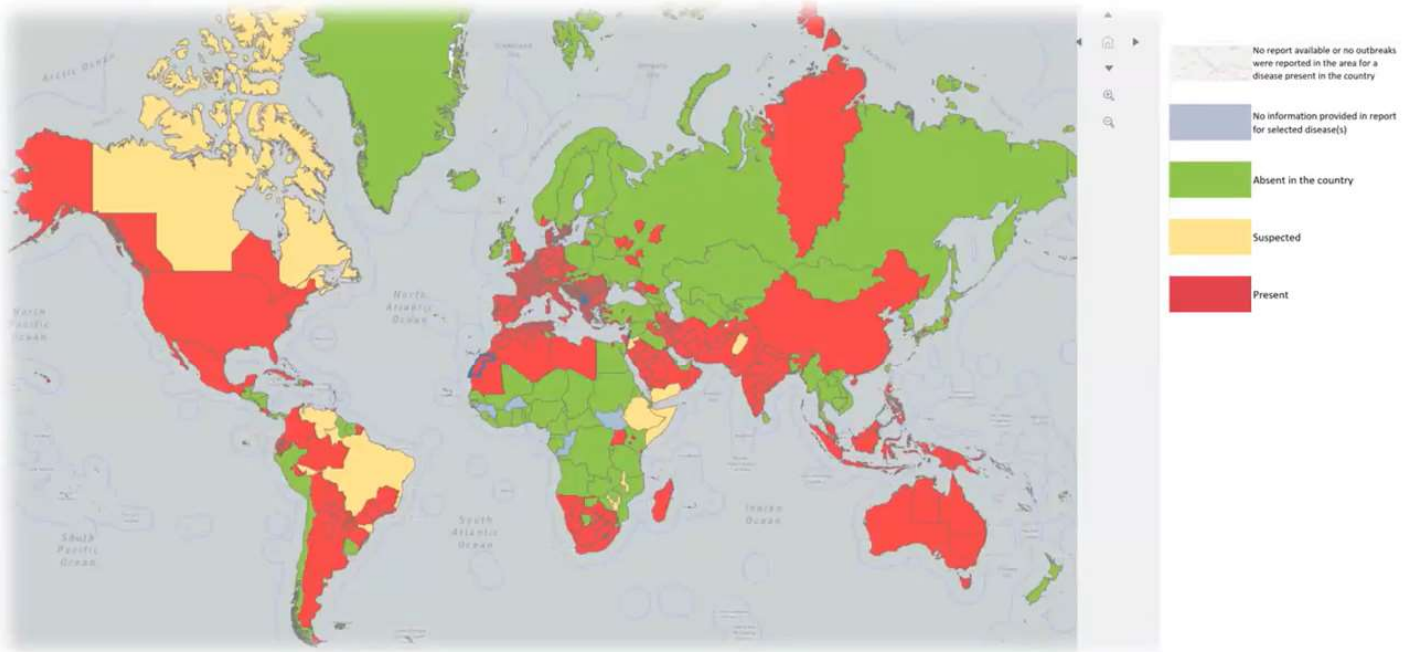




Clinical Signs

- Dependent on **host** (species, breed), **agent** (serotype, strain) and **environment**
- Virus-mediated vascular injury
 - Oedema, hyperaemia, haemorrhage
- Sheep *Morbidity up to 100% Mortality 0-30%*
 - fever, excessive salivation, depression, dyspnoea, panting
 - clear nasal discharge (later mucopurulent)
 - tongue swollen, cyanotic
 - erosions/ulcerations in mouth
- Cattle, Goats, Camelids *Morbidity < 5% and mortality rare*





Source: WAHIS. Accessed 16-09-24. <https://wahis.woah.org/#/dashboards/country-or-disease-dashboard>



<https://www.nvwa.nl/onderwerpen/dierziekten/documenten/dier/dierziekten/overige-dierziekten/publicaties/index>

BTV ecology is changing and the Australian surveillance program



France

- BTV-4 and BTV-8 are considered endemic in France.
- France has recently reported on the emergence of a new strain of BTV-8 that appears to be more pathogenic
- French authorities reporting that current BTV-8 vaccines are effective

French departments with reports of new BTV-8 strain (ESA: 381112023)



Figure 5: Map showing the departments in France affected by the recently emergent strain of BTV-8. Dark orange represents the department in which the new strain of BTV-8 was first reported, the lighter orange shows subsequent detections and the red line represents the SDEm coastal area where windborne incursion would be likely (This map was created from data published by Platform ESA, sourced at BIRVISA, accessed 29 November 2023).

Source: https://www.gov.uk/government/uploads/system/uploads/attachment_data/file/120468/2023_wildlife_outbreak_assessment_for_bluetongue_virus_in_europe.pdf



Spain

- BTV-1 and BTV-4 are endemic, but restricted to certain zones
- November 2023 – Outbreaks of BTV-4 reported in previously free regions of Madrid and Galicia (red dots)



Figure 7: Map showing 5 outbreaks of BTV-4 in Spain (red dots) reported 20 November 2023 by the Spanish Ministry of Agriculture and restriction zones for BTV-4, BTV-1 and unrestrictive regions (green) (Source: Spanish Ministry of Agriculture accessed on 29 November 2023).

Source: https://www.gov.uk/government/uploads/system/uploads/attachment_data/file/120468/2023_wildlife_outbreak_assessment_for_bluetongue_virus_in_europe.pdf



Europe – Historical situation (to 2018)

- Up until 1998, only sporadic BTV cases reported in Europe (Cyprus)
- 1998-2006 – Various BTV serotypes spread into Mediterranean regions and became established
- 2006 – BTV-8 outbreaks in Netherlands, Belgium, Germany, France and Luxembourg. Eradicated 2012 with mass vaccination.
- 2015 – Re-emergence of BTV-8 in France, subsequent detections in Switzerland, Germany, Belgium and Spain



Global Update - Europe – 2023-2024

- 2023/24 – Outbreaks of BTV-3 in North-Western Europe
- 2023 – Emergence of a new strain of BTV-8 in France
- Expansion of distribution of BTV distribution in Spain
- New outbreaks in Italy



Australian context

- Bluetongue first detected in Australia, 1975, Northern Territory
- Have since detected a total of **12 serotypes**: 1, 2, 3, 5, 7, 9, 12, 15, 16, 20, 21, 23 and multiple genotypes (Seg-3)
- Historically free from clinical disease, but has significant implications for animal export
- Present in NT, WA, QLD, NSW – with a seasonal distribution throughout
- *Culicoides brevitarsis* the primary vector (also *wadai*, *actoni*, *fulvis*)
- Two 'episystems' with significant serotype & genetic diversity in NT, northern WA; less diversity in eastern Australia



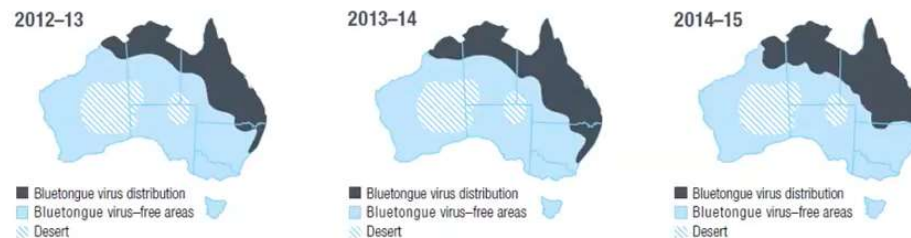


National Arbovirus Monitoring Program (NAMP)

- Monitors the distribution of three economically important arboviruses and their vectors: Bluetongue virus, Akabane virus, bovine ephemeral virus
- Jointly funded (industry: government)
- Multiple arbovirus surveillance strategies
- Objectives: market access, early warning, risk management



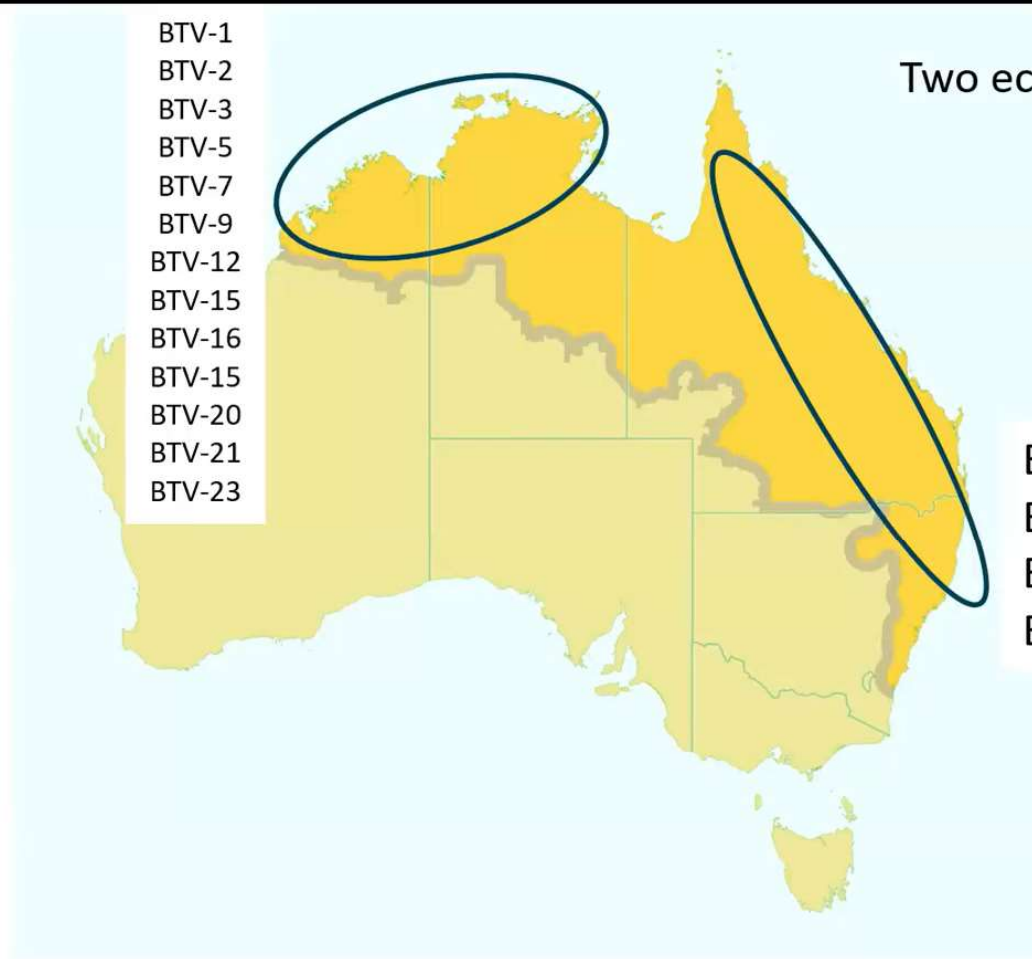
Figure 2 Distribution of bluetongue virus in Australia, 2012–13 to 2014–15



<https://animalhealthaustralia.com.au/maintaining-access-to-arbovirus-sensitive-markets/>



- BTV-1
- BTV-2
- BTV-3
- BTV-5
- BTV-7
- BTV-9
- BTV-12
- BTV-15
- BTV-16
- BTV-15
- BTV-20
- BTV-21
- BTV-23



Two ecozon

- BTV -1
- BTV-16
- BTV-15
- BTV-21

BTV-16 – introduced into NSW in 2017, BTV-15 Intermittent transmissions

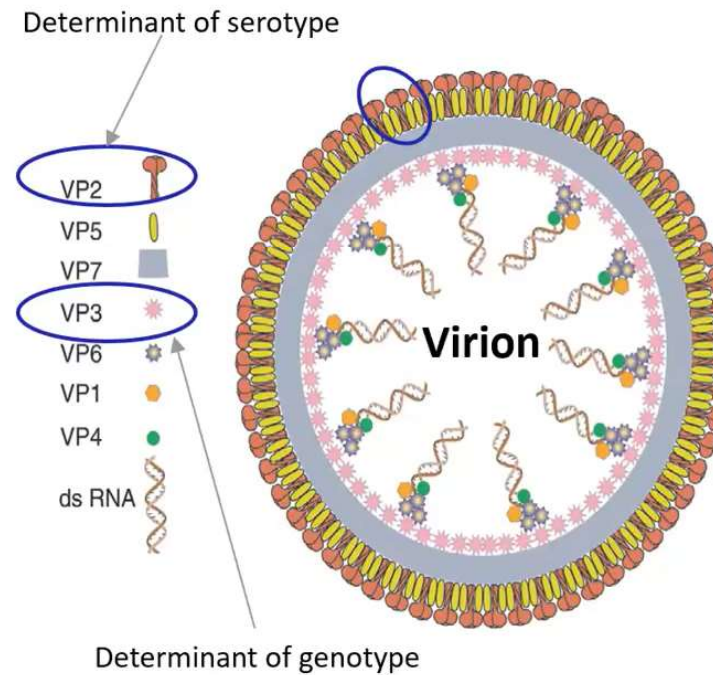


Diagnostic Testing & Advances @ ACDP



The virology

- Family *Reoviridae*; genus *Orbivirus*
 - *Related to African Horse Sickness virus*
- ds RNA segmented genome
- 28⁺ serotypes
- **Segmented Genome – 10 distinct segments. Gene reassortment known to occur.**

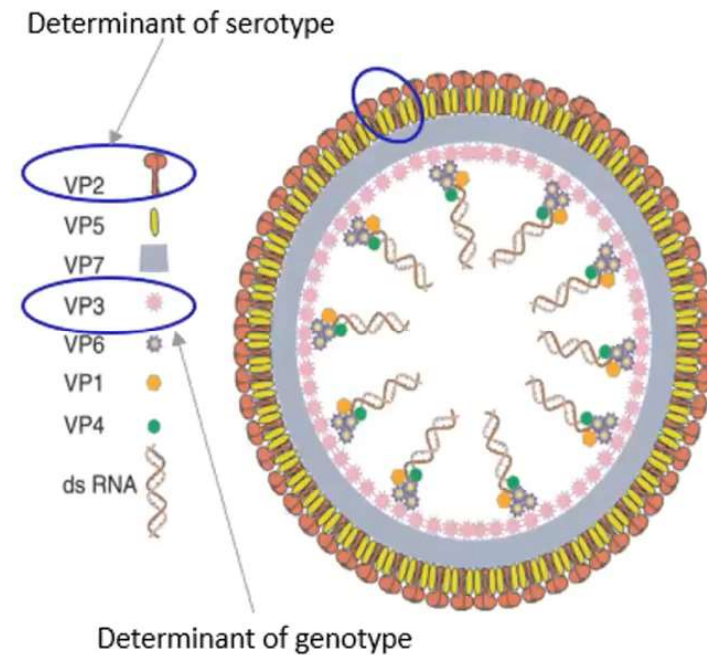




Gaps & Opportunities

Traditional testing does not:

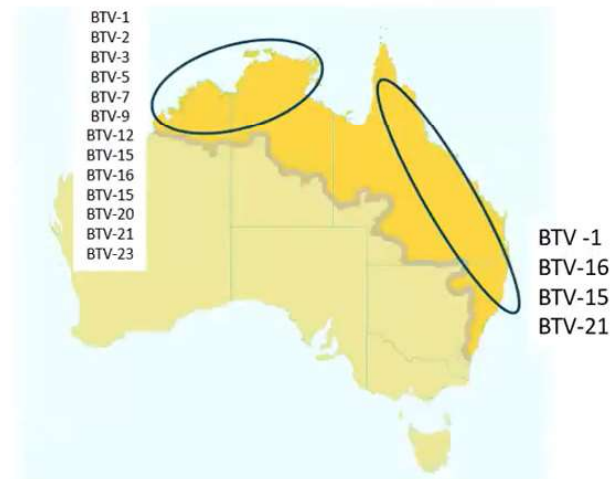
- Inform a detailed understanding of transmission networks, as it is focussed on only 2/10 gene segments
- Provide spatio-temporal context
- Consider the role of other gene segments in the epidemiology, and potentially, pathogenicity of BTV
 - **Virulence determinants unknown – likely associated with multiple gene segments**





Recent developments at ACDP - genomics

- Developed a model that can perform fine-grained characterization of the genetic relationships between BTV isolates **at the whole genome level**.
 - To better understand the relationships between isolates
 - Spatio-temporal within Australia and outside of our country borders
 - Potential future application to identifying virulence determinants
- Can visualise this information (along with spatiotemporal data) in Nextstrain®



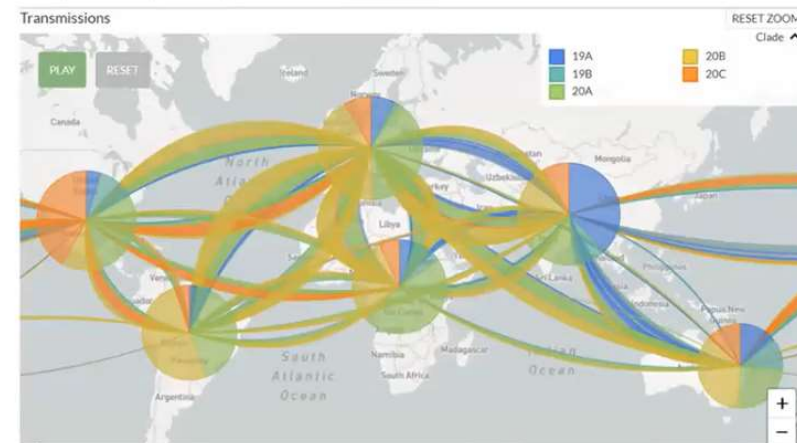


Nextstrain : now being applied for BTV in Australia

Genomic epidemiology of novel coronavirus - Global subsampling

Maintained by the Nextstrain team. Enabled by data from [GISAID](#)

Showing 3743 of 3743 genomes sampled between Dec 2019 and Oct 2020.



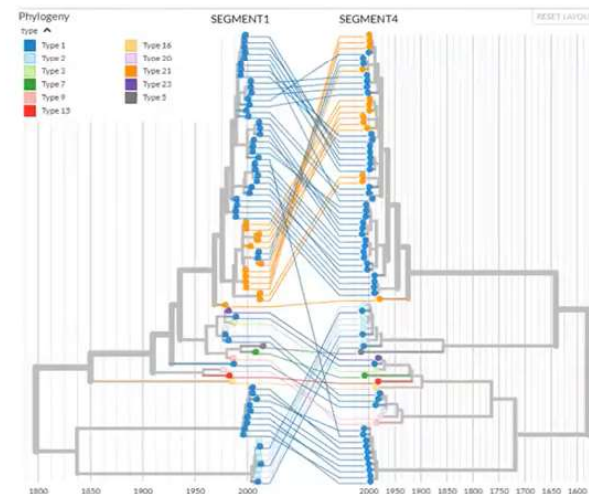
Map of possible global transmission pattern of SARS-CoV-2 clades from a subset of GISAID whole genome sequencing data.

Source: Nextstrain



Whole Genome Sequencing and Nextstrain

- Greatly improves our understanding of the molecular epidemiology of BTV
- Improved understanding of relationships between viruses to **understand changes in virus transmission**
- Improved ability to detect:
 - **recombination events**
 - **incursion & spread of exotic gene segments**





Thank you

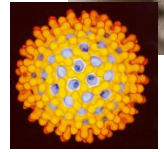
Dr Stacey Lynch
Team Leader, Mammalian
Infectious Disease Research

Stacey.Lynch@CSIRO.au

**On behalf of the
WOAH Reference Laboratory for
Bluetongue virus**

Australia's National Science Agency





EMERGENCE OF EPIZOOTIC HEMORRHAGIC DISEASE (EHD) IN EUROPE

STEPHAN ZIENTARA

ANSES - DIRECTOR OF ANIMAL HEALTH LABORATORY, FRANCE

EU/WOAH/FAO REFERENCE LAB ON FMD
EU REFERENCE LAB ON EQUINE DISEASES
WOAH REFERENCE LAB ON EHDV



Epizootic Haemorrhagic disease





Virology

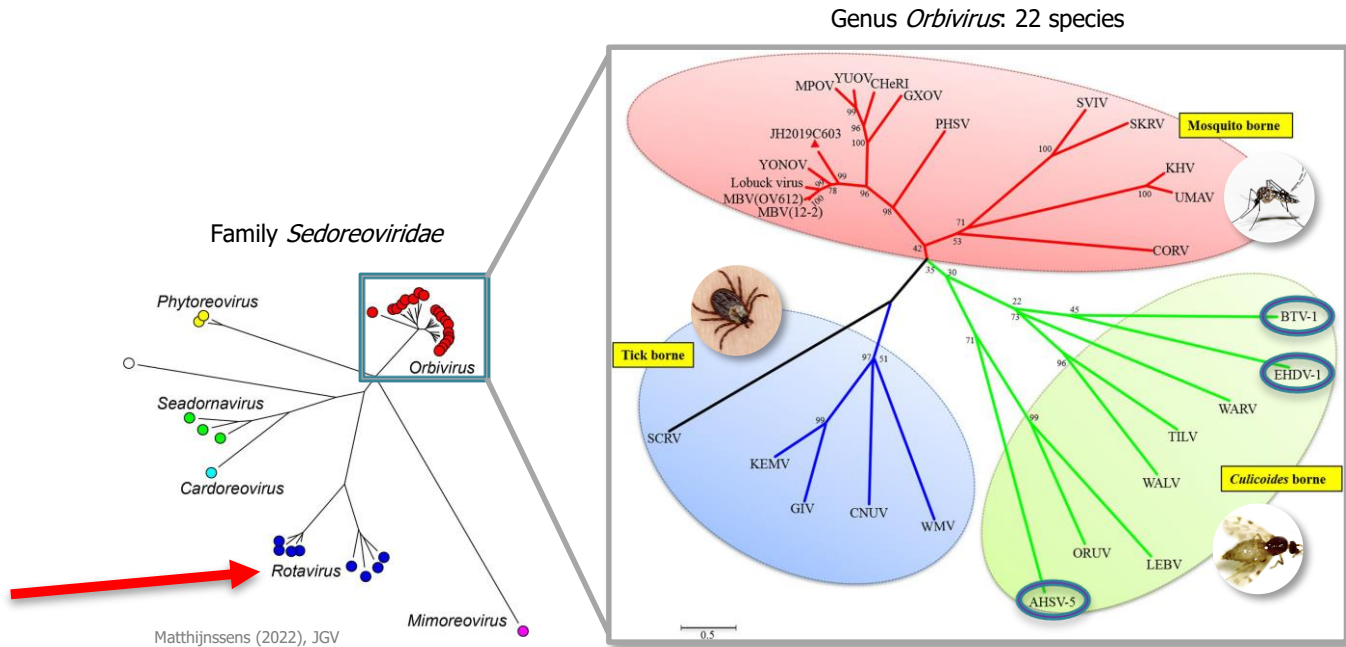
Disease

Epidemiology

Diagnosis

Prevention

Orbivirus



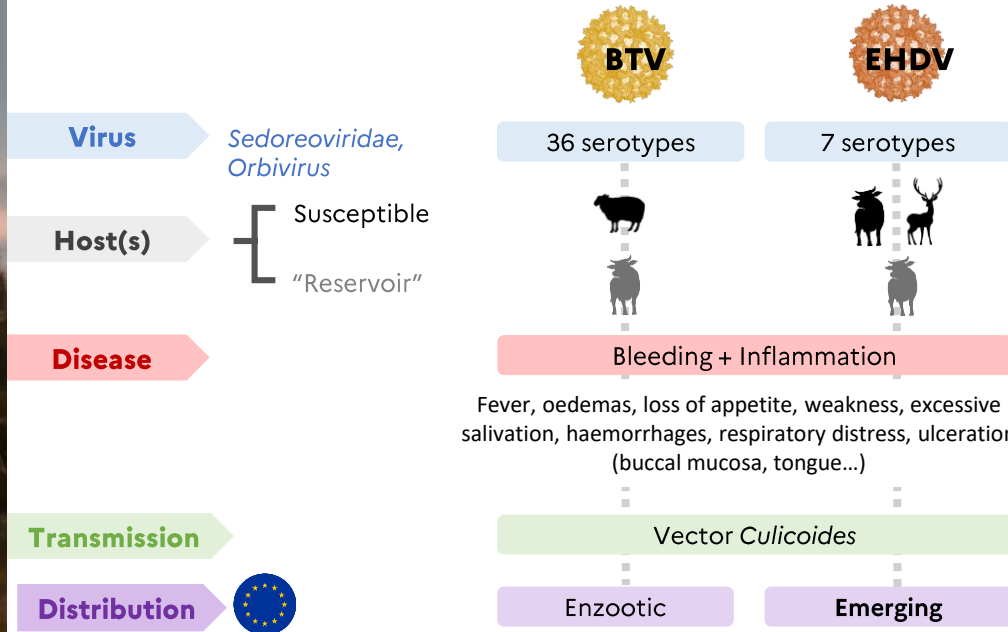
3 important diseases

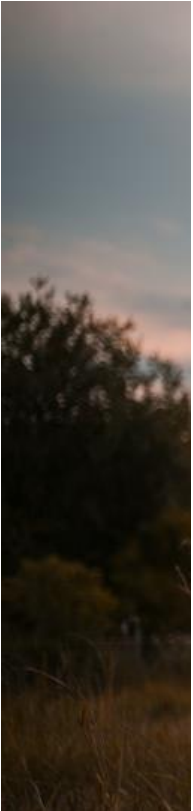
- **BTV:** Bluetongue virus
- **EHDV:** Epizootic hemorrhagic disease virus
- **AHSV:** African horse sickness virus



Bluetongue (BTV) & Epizootic Hemorrhagic Disease (EHD)

WOAH & European notifiable animal diseases





Sedoreoviridae, Orbivirus

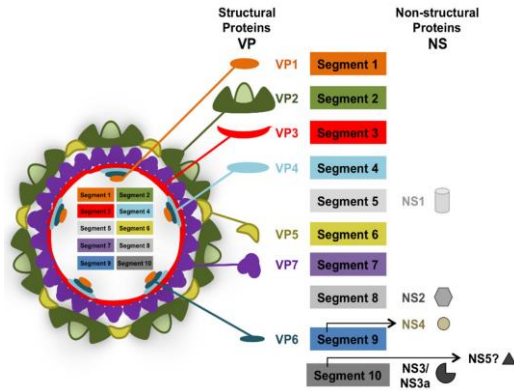
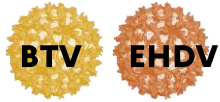
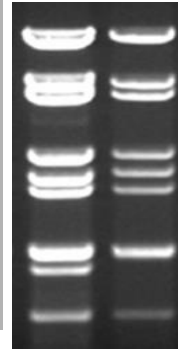
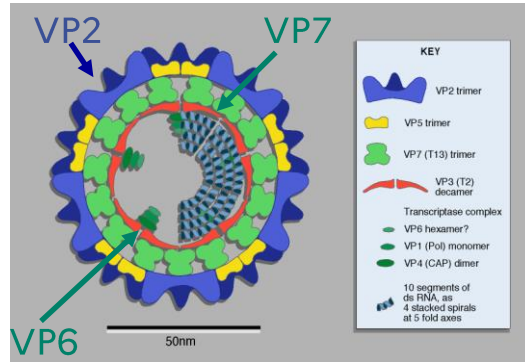


Figure from Rodriguez-Martin *et al.*, 2021

- Non-enveloped segmented dsRNA+ genome
- 10 segments \approx 19,2 kb
- Conserved sequence at the extremities:
5' [GUU(A/U)A(A/U) ... AC(A/U)UAC] 3'
- S1 to S8: Monocistronic segment
- S9 & S10: Bicistronic segments
- **Serotyping: Segment 2**

reassortment

EHDV caused by an Orbivirus Familie : *Sedoreoviridae*



7 serotypes

Reference strains						
EHDV-1 USA1955/0	EHDV-2 CAN1962/01	EHDV-3 [M.Dona1] NIG1967/0	EHDV-4 NIG1968/01	EHDV-5 AUS1977/01	EHDV-6 AUS1981/07	EHDV-7 AUS1981/06
	EHDV-2 (Ibaraki virus) JAP1959/01					EHDV-8 AUS1982/06

Table 3: Commonly accepted reference strains in the ds RNA virus collection at Institute for Animal Health (IAH) Pirbright, UK and at the Arthropod-Borne Animal Diseases Research Laboratory (ABADRL), USA.

New serotypes?

South Africa



EHDV-9?

Wright, I. M. 2013: *Serological and Genetic Characterisation of Putative New Serotypes of Bluetongue Virus and Epizootic Haemorrhagic Disease Virus Isolated From an Alpaca. North-West University, South Africa*

Japan (1998)



EHDV-10?



China



EHDV-11 ?

> Emerg Infect Dis. 2020 Dec26(12):3081–3083. doi: 10.3201/eid2612.191301.

Novel Serotype of Epizootic Hemorrhagic Disease Virus, China

Heng Yang, Zhuoran Li, Jinping Wang, Zhanhong Li, Zhenxing Yang, Defang Liao, Jianbo Zhu, Huachun Li

PMID: 33219797 PMCID: PMC7706924 DOI: 10.3201/eid2612.191301

Although recognised earlier in the south-eastern United States,

EHD was first described after a severe outbreak of the disease in **white-tailed deer** (*Odocoileus virginianus*)
in New Jersey in 1955

Shope R.E., Macnamara L.G. & Mangold R. (1960). – A virus-induced epizootic hemorrhagic disease of the Virginia white-tailed deer (Odocoileus virginianus). J. Experim. Med., 111, 155–170



Review

Epizootic haemorrhagic disease N.J. Maclachlan, S. Zientara, G. Savini & P.W. Daniels

Rev. Sci. Tech. Off. Int. Epiz., 2015, 34 (2), 341-351

Richard Edwin Shope

- **Richard Edwin Shope (1901-1966)**



- **1924:** instructor in pharmacology at the University of Iowa, studying **tuberculosis**, and rapidly joined the laboratories of the **Rockefeller Institute** at **Princeton**.



- **1928:** he left the field of tuberculosis to work on **hog cholera** and thus began a career in the field of **virology**.
- **1931:** identified **swine influenza**, together with his mentor Paul A. Lewis.
- **1932:** rabbit **fibroma**.
- **1933:** rabbit **papilloma**.

"A new disease of deer"

...but

A VIRUS-INDUCED EPIZOOTIC HEMORRHAGIC DISEASE OF
THE VIRGINIA WHITE-TAILED DEER
(ODOCOILEUS VIRGINIANUS)*
BY RICHARD E. SHOPE, M.D., LESTER G. MACNAMARA, AND
ROBERT MANGOLD
*(From The Rockefeller Institute; and State of New Jersey Department of Conservation
and Economic Development, Division of Fish and Game, Trenton)*
PLATES 7 TO 10
(Received for publication, September 22, 1959)

Shope RE (1960, J Exp Med)

"Among woodsmen and hunters in the South, the disease has long borne the name "**black tongue**," presumably because one of the characteristics of a deer dead of it was a discolored and swollen tongue which generally protruded from the mouth. Ruff...has given a good description of "black tongue" (*Ruff FJ ; 1950. Wild Life in North Carolina*).

WHAT IS "Black Tongue" AMONG DEER?

BY FRED A. RUFF*

"we had been in **error** in our belief. Search of the **files** of the **United States Forest Service**... outbreaks of a fatal epizootic disease similar to the one we encountered in New Jersey have occurred in deer in various of the **southeastern states** at irregular intervals at least since **1890***..."

"However, because this name has confusing connotations... we propose, therefore, to continue to designate the condition as **epizootic hemorrhagic disease of deer (EHD)**."

*Leonard E. Foote (Wild Life Management Institute; personal communication)

First reported epizootics of EHD



Spread from separated foci during 8-10 w.

Late summer/early fall (terminated before winter).

Damien Vitour



Ibaraki strain EHDV2
Isolated from a cattle in 1959 in Ibaraki, Japan

Ibaraki



Virology

Disease

Epidemiology

Diagnosis

Prevention

Why Epizootic Haemorrhagic Disease of **DEER** ?





Transmission by
Culicoides sonorensis



other species

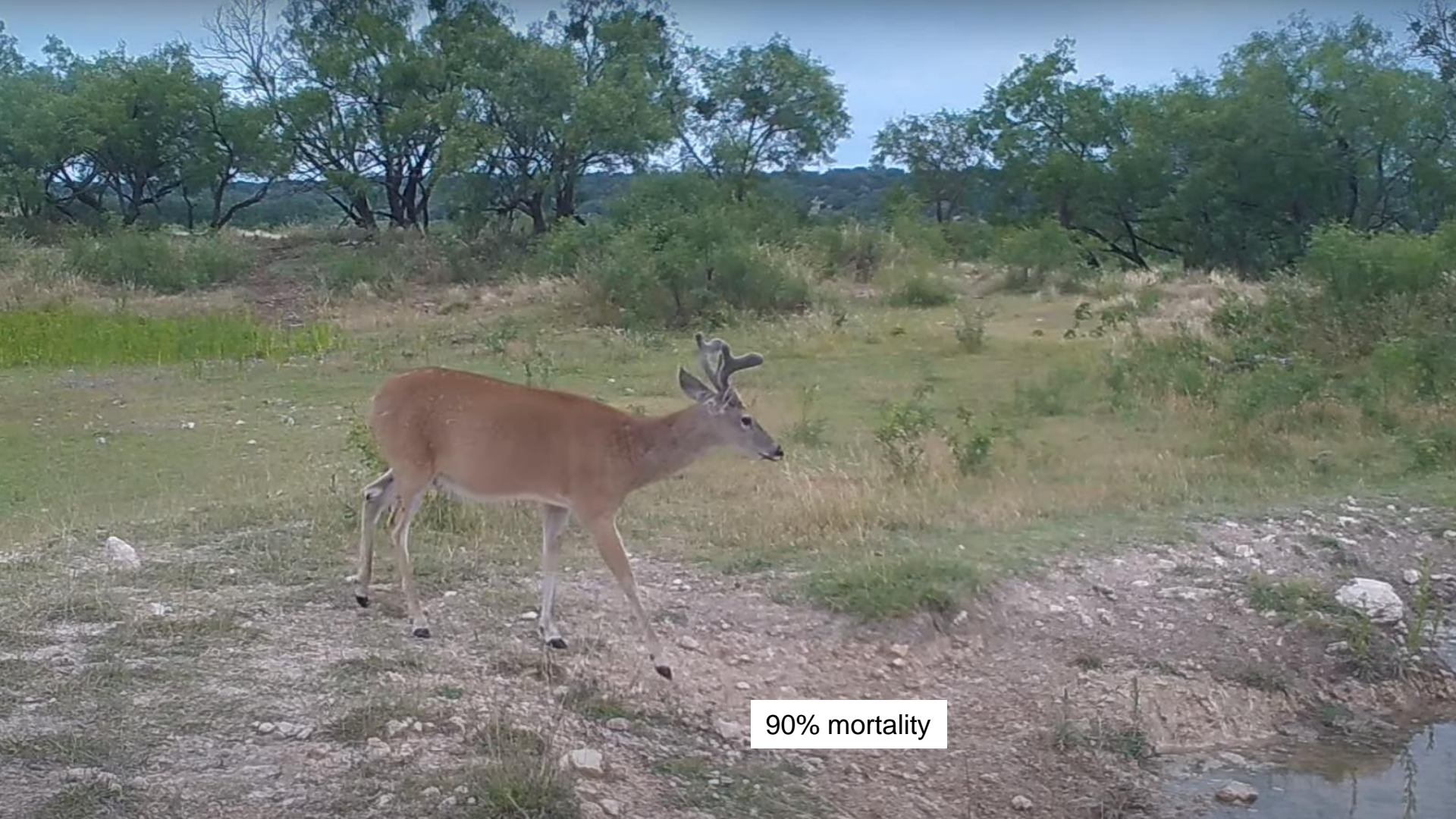
- C. insignis*
- C. mohave*
- C. debilipalpis*
- C. obsoletus*
- C. scoticus*
- C. paraensis*
- C. spinosus*
- C. stellifer*

...



Disease Transmission Cycle of Epizootic Hemorrhagic Disease (EHD)





90% mortality

- EHDV serotypes **1, 2, 6, 7 and 8** induce clinical signs in cattle

Epizootic haemorrhagic disease virus (EHDV) mainly infects deer, but sheep and cattle can also be infected.

EHDV added to the WOAHL list in **May 2008.**

- Loss of appetite
- Fear of humans lost
- Extensive haemorrhages
- Weakness
- Excessive salivation
- Rapid pulse and respiratory rate
- Fever
- Blue tongue from lack of oxygenated blood
- Breaking of hooves caused by growth interruptions
- Diarrhoea
- Unconsciousness
- Death



Virology

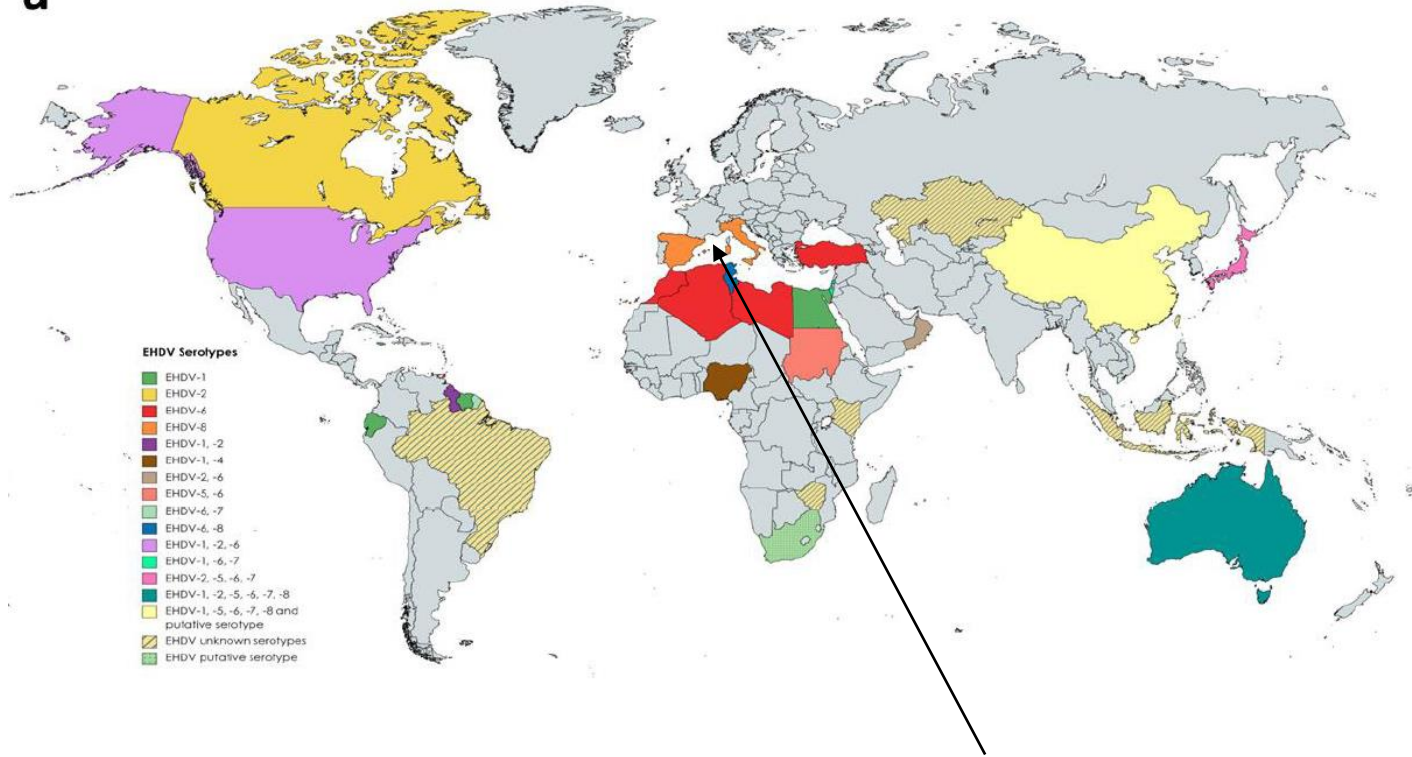
Disease

Epidemiology

Diagnosis

Prevention

a






microorganisms

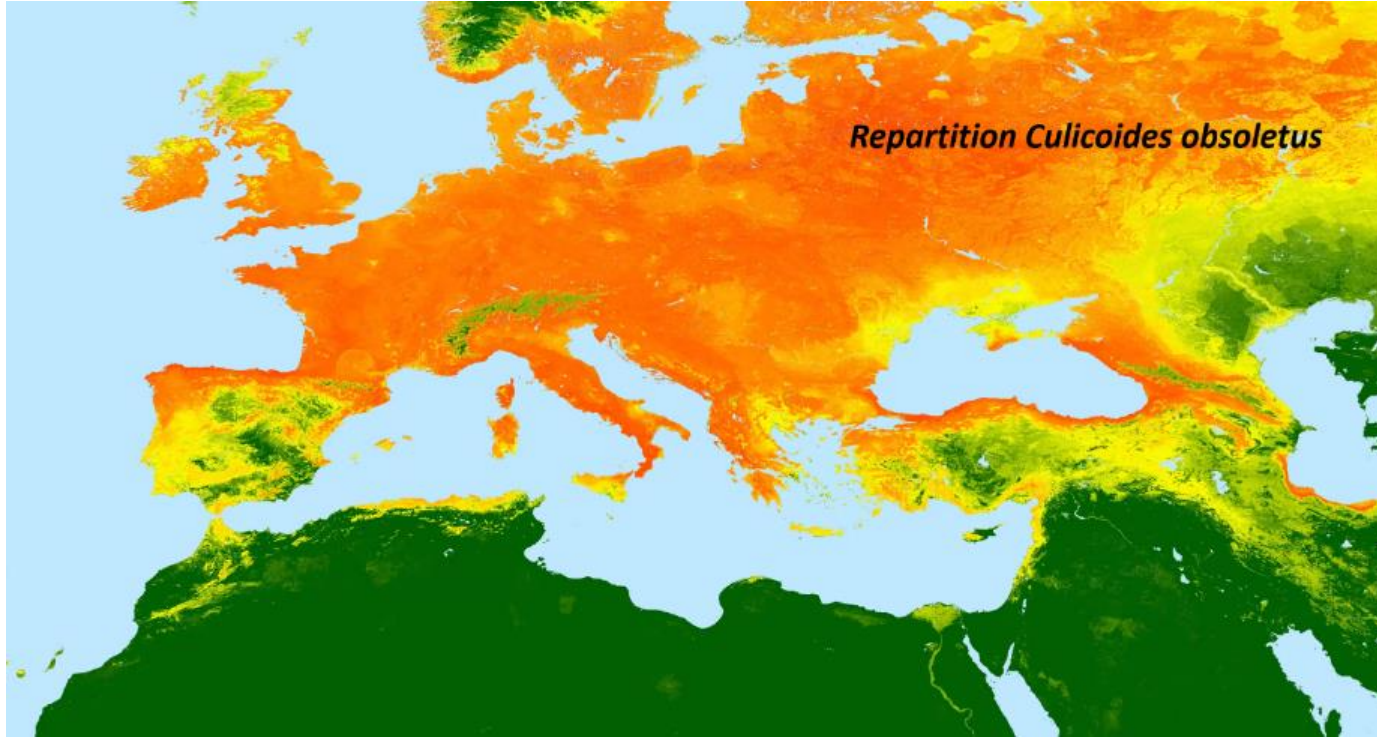


2022 ; 2023

Review

Epizootic Hemorrhagic Disease Virus: Current Knowledge and Emerging Perspectives

Luis Jiménez-Cabello , Sergio Utrilla-Trigo, Gema Lorenzo, Javier Ortega  and Eva Calvo-Pinilla 



EHDV-8



Epizootic Haemorrhagic Disease virus serotype 8 in Tunisia, 2021

2021

Soufien Sghaier¹, Corinne Sailleau², Maurilia Marcacci³, Sarah Thabet¹, Valentina Curini³, Thameur Ben Hassine⁴, Liana Teodori³, Ottavio Portanti³, Salah Hammami⁵, Lucija Jurisic^{3,6}, Massimo Spedicato³, Lydie Postic², Ines Gazani⁷, Raja Ben Osman⁸, Stephan Zientara², Emmanuel Breard², Paolo Calistri³, Juergen A. Richt⁹, Edward C. Holmes¹⁰, Giovanni Savini³, Francesca Di Giallonardo¹¹, and Alessio Lorusso^{3*}



Figure 1. Clinical signs in cattle. (A) Teat erosions, (B) Oral congestion and erosions, (C) Submandibular oedema, conjunctivitis, and lacrimation, (D) Nasal discharge and mucosal erosion.

200 outbreaks

2022

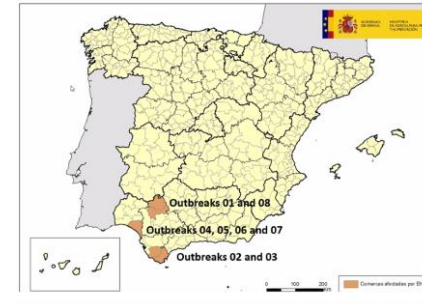


From 28 Octobre 2022

Clinical signs

- Cattle and a deer in Sardinia and Sicily (morbidity rate 5 to 10 %)
- Cattle in Andalusia (morbidity rate 10 to 15 % ; mortality rate <1%)

➔ Confirmation EHDV-8



EMERGING INFECTIOUS DISEASES®

EID Journal > Volume 29 > Number 5—May 2023 > Main Article

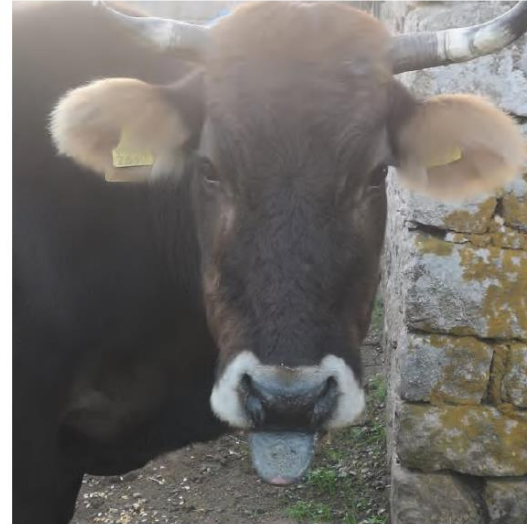
Volume 29, Number 5—May 2023

Research Letter

Epizootic Hemorrhagic Disease Virus Serotype 8, Italy, 2022

Alessio Lorusso¹, Stefano Cappai, Federica Loi, Luigia Pinna, Angelo Ruiu, Giontonella Puggioni, Annalisa Guercio, Giuseppa Purpari, Domenico Vicari, Soufien Sghaier, Stephan Zientara, Massimo Spedicato, Salah Hammami, Thameur Ben Hassine, Ottavio Portanti, Emmanuel Breard, Corinne Sailleu, Massimo Ancora, Daria Di Sabatino, Daniela Morelli, Paolo Calistri, and Giovanni Savini

The first infected animal



Alessio Lorusso, IZST

2023



18 September 2023

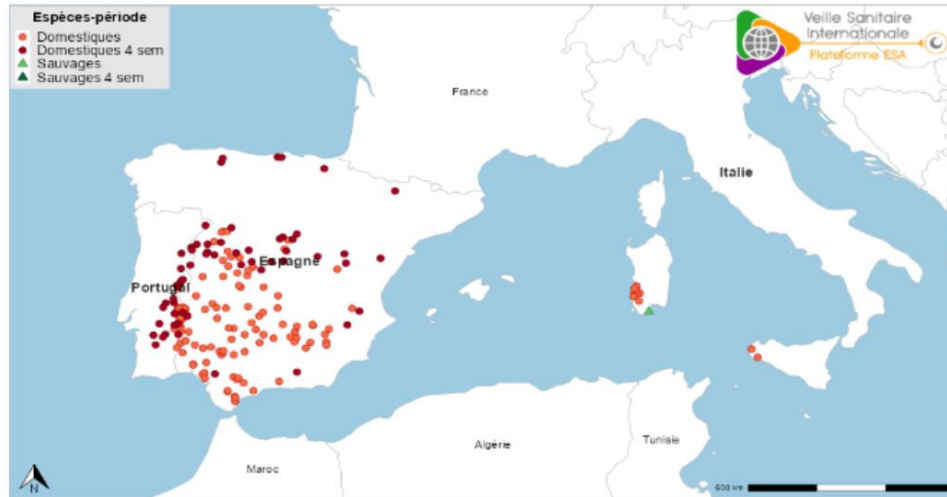
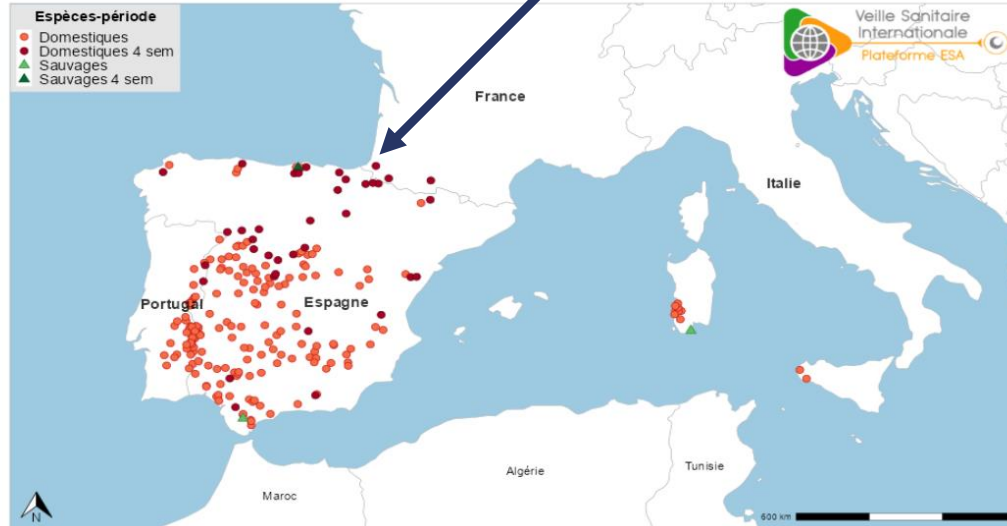


Figure 1. Localisation

September 2023



Hautes-Pyrénées : détection d'un cas de maladie hémorragique épizootique (MHE) dans un élevage bovin



Pyrénées Atlantiques (2), Hautes Pyrénées (1)

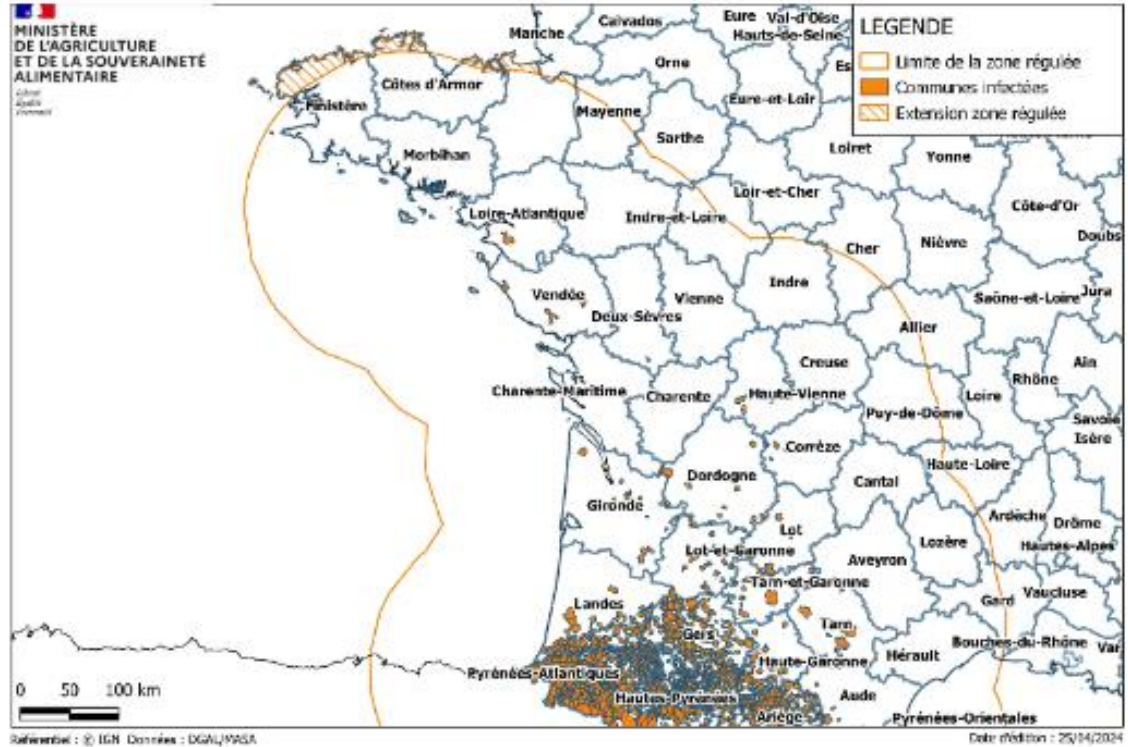
18/09/2023



> 4 330 outbreaks

20 infected departments:
09, 11, 19, 24, 31, 32, 33, 40,
46, 44,
47, 64, 65, 81, 82, 85, 87

1 izard
2 deers
1 roe deer



> [Virologie \(Montrouge\)](#). 2024 Feb 1;28(1):1-2. doi: 10.1684/vir.2024.1035.

Emergence of Epizootic Hemorrhagic Disease in France in 2023: Impacts and Future Prospects

Stéphan Zientara, Corinne Sailleau, Pascal Dujardin, Emmanuel Bréard, Damien Vitour

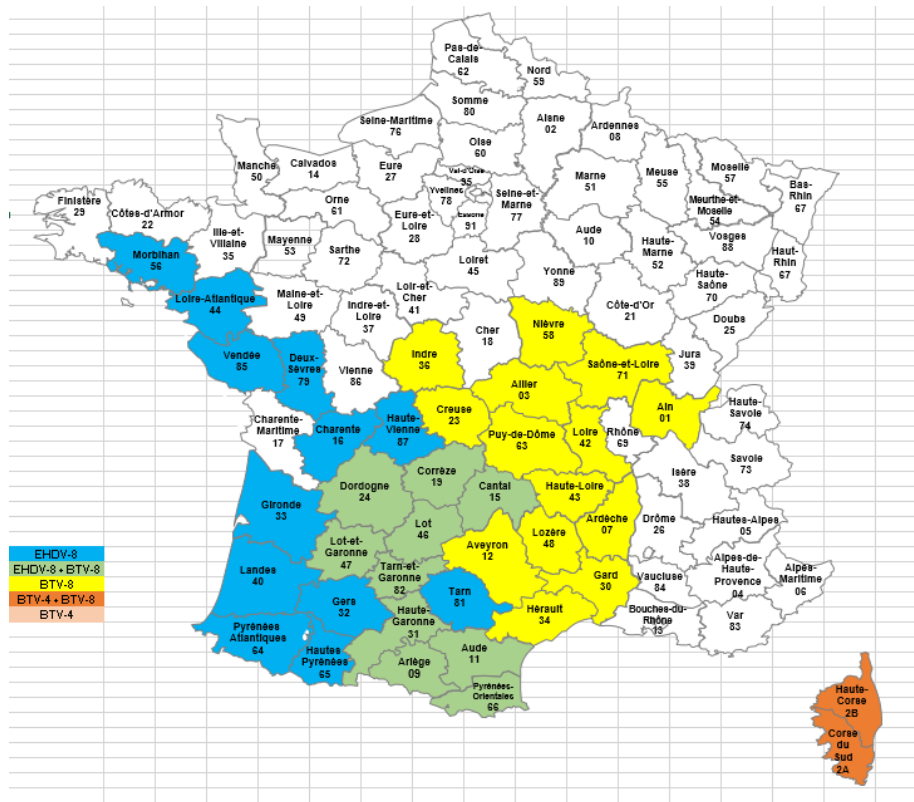


Dr Vivien Philis, Lannemezan
Dr Mylène Lemaire-Meyer (LVD09)



Dr Alberto Jorda Blanco (Aude)
Dr Mylène Lemaire-Meyer (LVD09)

29/01/24







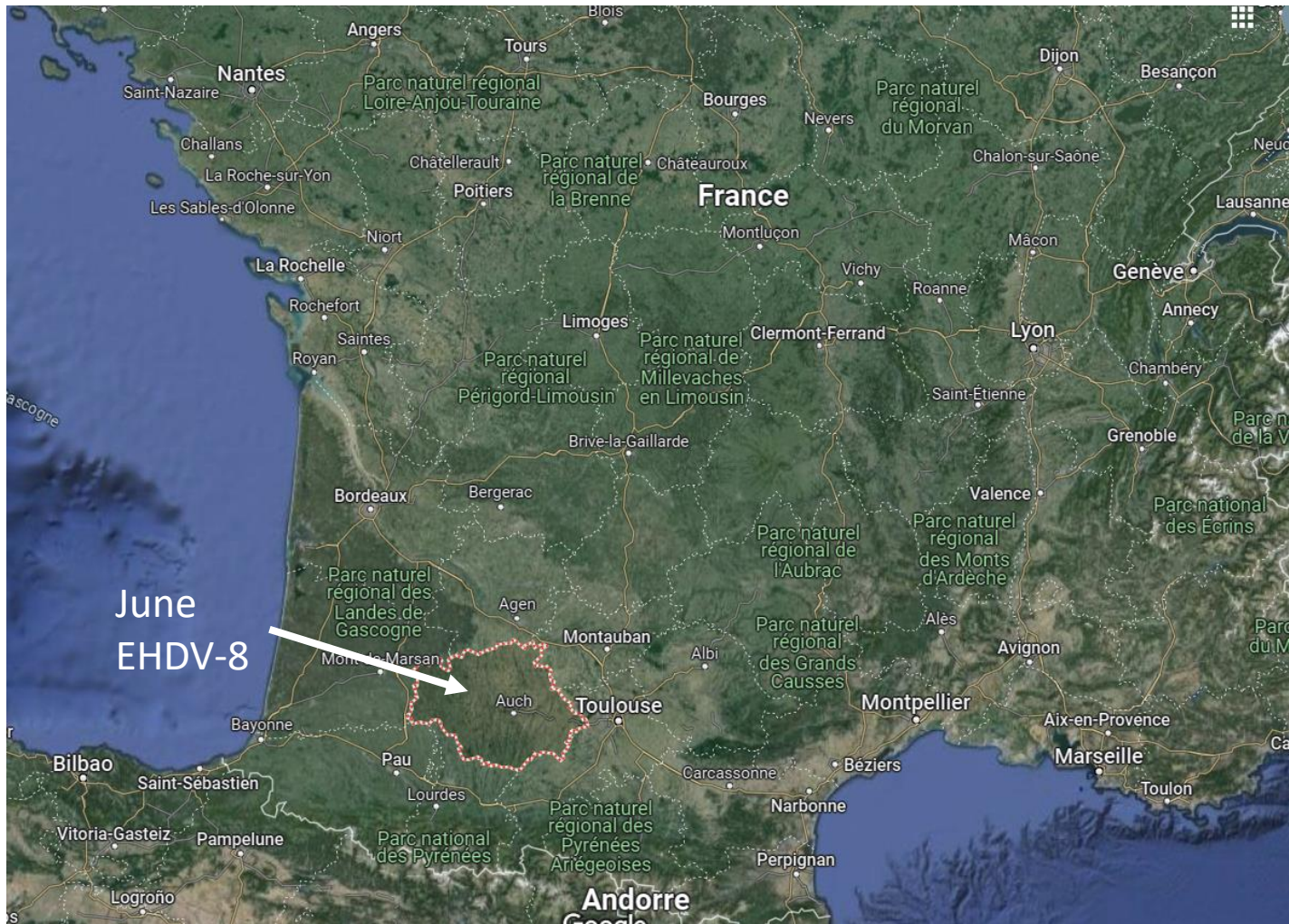
June 2024

Confirmado el primer caso de enfermedad hemorrágica epizootica de la actual temporada vectorial

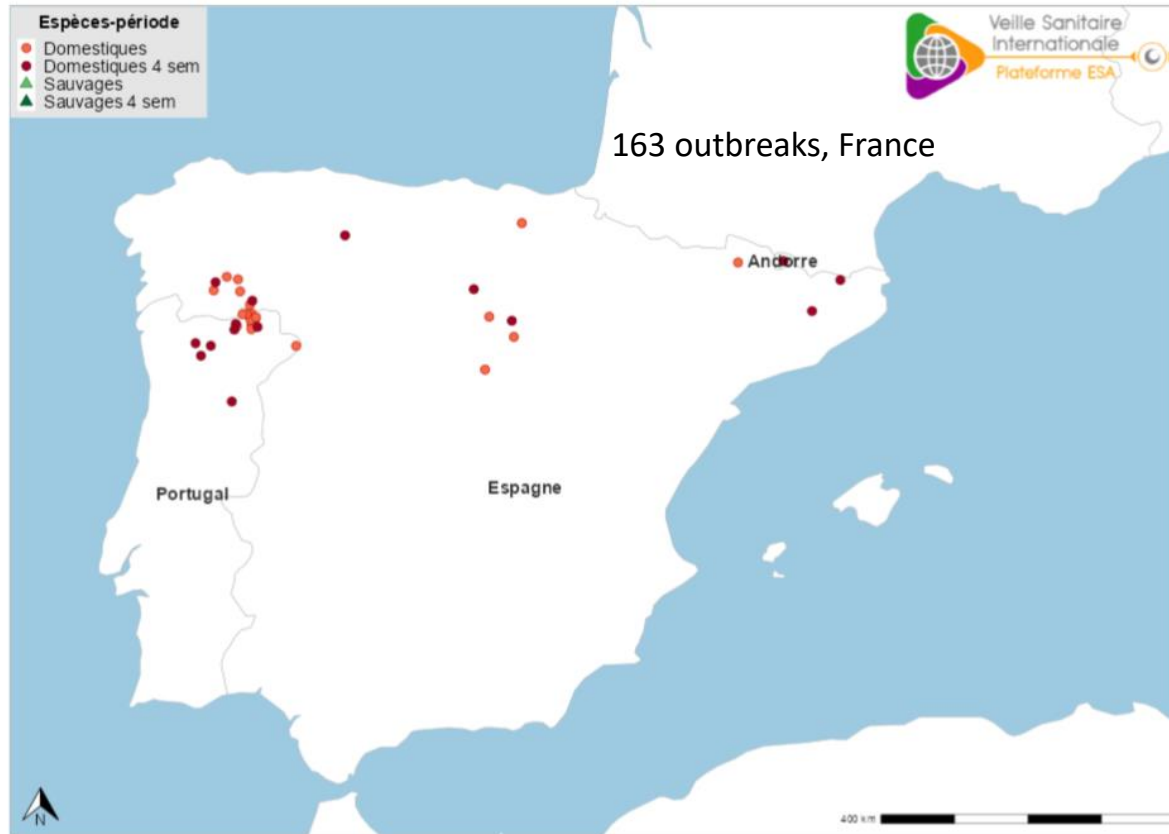
El Ministerio de Agricultura, Pesca y Alimentación ha confirmado el primer caso de enfermedad hemorrágica epizootica en una explotación de bovino de Guadalajara



Vacas libres de enfermedad hemorrágica epizootica según la última actualización del Gobierno. (Foto: Freepix)



EHD
18 August 2024





Virology

Disease

Epidemiology

Diagnosis

Prevention

> [Vet Ital.](#) 2023 Dec 20. doi: 10.12834/VetIt.3433.23112.1. Online ahead of print.

Experimental infection of cattle, sheep, and goats with the newly emerged epizootic hemorrhagic disease virus serotype 8

Massimo Spedicato ¹, Francesca Profeta ¹, Sarah Thabet ², Liana Teodori ¹, Alessandra Leone ¹, Ottavio Portanti ¹, Maura Piscicella ¹, Barbara Bonfini ¹, Simone Pulsoni ¹, Francesca Rosso ¹, Emanuela Rossi ¹, Paola Ripà ¹, Angela De Rosa ¹, Eugenia Ciarrocchi ¹, Roberta Irelli ¹, Antonio Cocco ¹, Corinne Sailleu ³, Nicola Ferri ¹, Tiziana Di Febo ¹, Damien Vitour ³, Emmanuel Breard ³, Daniele Giansante ¹, Soufien Sghaier ⁴, Thameur Ben Hassine ⁵, Stephan Zientara ³, Romolo Salini ¹, Salah Hammami ², Giovanni Savini ¹, Alessio Lorusso ⁶

Affiliations + expand

PMID: 38117055 DOI: [10.12834/VetIt.3433.23112.1](#)



A



B



C

6 calves, 4-5 months old, 2 males ; 4 females
Strain EHDV-8, Sardinia 2022, 1 passage BSR cells
2 ml inoculum SC+ 1 ml ID, 6,7 log₁₀ TCID₅₀/ml

3/6: fever
1 calf: clinical signs

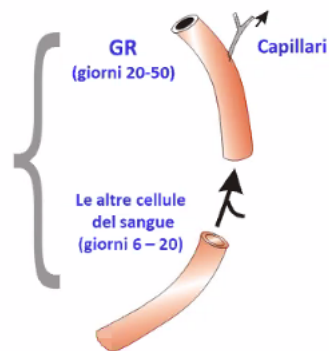
Viraemia

BT

- Cattle
 - VI: 5-60 dd
 - RT-PCR: 5-180 dd
- Sheep
 - VI: 5-30 dd
 - RT-PCR: 5-100 dd
- Goats
 - VI: 5-30 dd
 - RT-PCR: 5-100 dd

EHD

- Cattle
 - VI: 7-42 dd (EHDV-8: 2-16 dd)
 - RT-PCR: 2-180 dd
- Sheep
 - VI: 2-7
 - RT-PCR: 5-100 dd
- Goats
 - VI: 0
 - RT-PCR: 0





Laboratoire de santé animale
De Maisons Alfort
LNR Fièvre catarrhale ovine et
MHE

10/04/2024

Validated EHDV PCR

	KITS	FOURNISSEURS
Dépistage de groupe « tout génotype » (RT-PCR duplex)	Méthode interne (LNR)	
	ID Gene™ EHDV Duplex (id VET)	ID
	VETMAX™ EHDV	Life Technologies SAS
	Bio-T kit® EHDV	BioSellal
	ADIAVET™ EHDV REAL TIME	BioX
	ID Gene™ EHDV Advantage Duplex	ID

Inactivated vaccine EHDV-8

Temporay authorization in France

Cattle

2 injections primo-vaccination.

Prevention of viremia (PCR >40) and reduction of clinical signs



Ceva Animal Health

➤ 190 outbreaks

22 August 2024





Acknowledgements



Corinne Sailleau



Giovanni Savini



Alessio Lorusso



Lydie Postic



Mathilde Turpaud



Damien Vitour



Emmanuel Bréard

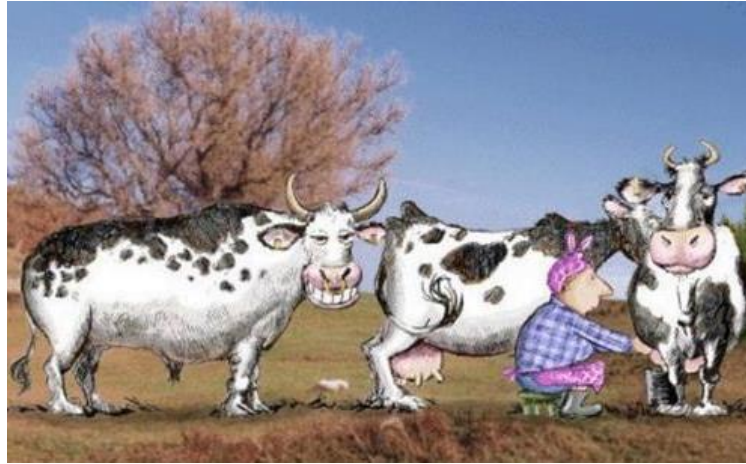


Mathilde Gondard



Soufien Sghaier

Thank you for your attention



WOAH Regional Workshop
on Vector Borne diseases in
Asia and the Pacific

19-20 September 2024

LUMPY SKIN DISEASE

WOAH MANUAL: CHAPTER 3.4.12.

Antoinette van Schalkwyk

Agricultural Research Council – Onderstepoort Veterinary Institute

Lumpy skin disease: Introduction



Causative agent: [Poxvirus] – {*Capripoxvirus*}
- Lumpy skin disease virus

Type strain: Neethling strain (Western Cape, RSA, 1957)

Ruminants: Cattle and water buffalo.

Wildlife: Springbok, impala, eland, giraffe, camel, gazelle, oryx, Arabian oryx, banteng and Mainland searow

Notifiable disease: WOAH

Lumpy skin disease: Transmission

Highly contagious
viral infection

Direct contact: Skin nodules, nasal discharge or saliva from infected animals

In-direct: Contaminated food, water and milk

Vertically: Intrauterine route

Vector-borne,
non-zoonotic and
transboundary
disease

Long-distance dispersal of LSDV seems to occur via the movement of infected animals

NOT an arbovirus

Seasonal patterns indicate that arthropod-borne transmission (Mechanical):

Blood-sucking or biting arthropods:

Stable flies (*Stomoxys calcitrans*), Mosquitoes (*Aedes aegypti*),

Hard ticks (*Rhipicephalus* and *Amblyomma* species)

House fly (*Musca domestica*)

Require more
investigations

Lumpy skin disease: Role of wildlife species

This evidence is supported by extensive serosurveys of wildlife in Africa which detected antibodies in very low numbers of samples (Davies, 1982; Hedger and Hamblin, 1983), leading the authors to conclude “wildlife in Africa probably does not play a very important part in the perpetuation and spread of LSDV”.

LSDV has spread rapidly in recent years into the Middle East and Asia, and the susceptibility of wildlife species in these regions to LSDV is unknown.



In South Africa:
Springbuck and giraffe - National
parks and game reserves

Require more investigations

In 2022, isolation and characterisation of LSDV from a giraffe in a zoo was reported in Vietnam (Dao et al., 2022).

Namibia: an eland antelope (*Taurotragus oryx*), which was asymptomatic for LSD, but LSDV DNA isolated (Molini et al., 2021)

India: Farmed camels (Kumar et al., 2023)

India: Free living gazelle (Sundhankar et al., 2023)

Lumpy skin disease: Global spread

Zambia: 1929

South Africa: 1944

Kenya: 1958

Middle East: 1988
and again in 2006

Europe, Caucasus and
Russia: 2015

China and India:
2019



Lumpy skin disease: Epidemiology

Clusters based on Markers



- 1.2
- 1.2 -KSGP
- 1.1 & 1.2
- 1.2 & 2.3
- 2.5
- 1.1 & 1.2 & 2.1 & 2.2 & 2.4 & 2.5 & 2.6
- No sequence data

Lumpy skin disease: Phylogenetics

Clusters 1.1 and 1.2

LSDVs representing sequences from both clusters were circulating in Africa in the 1950's.

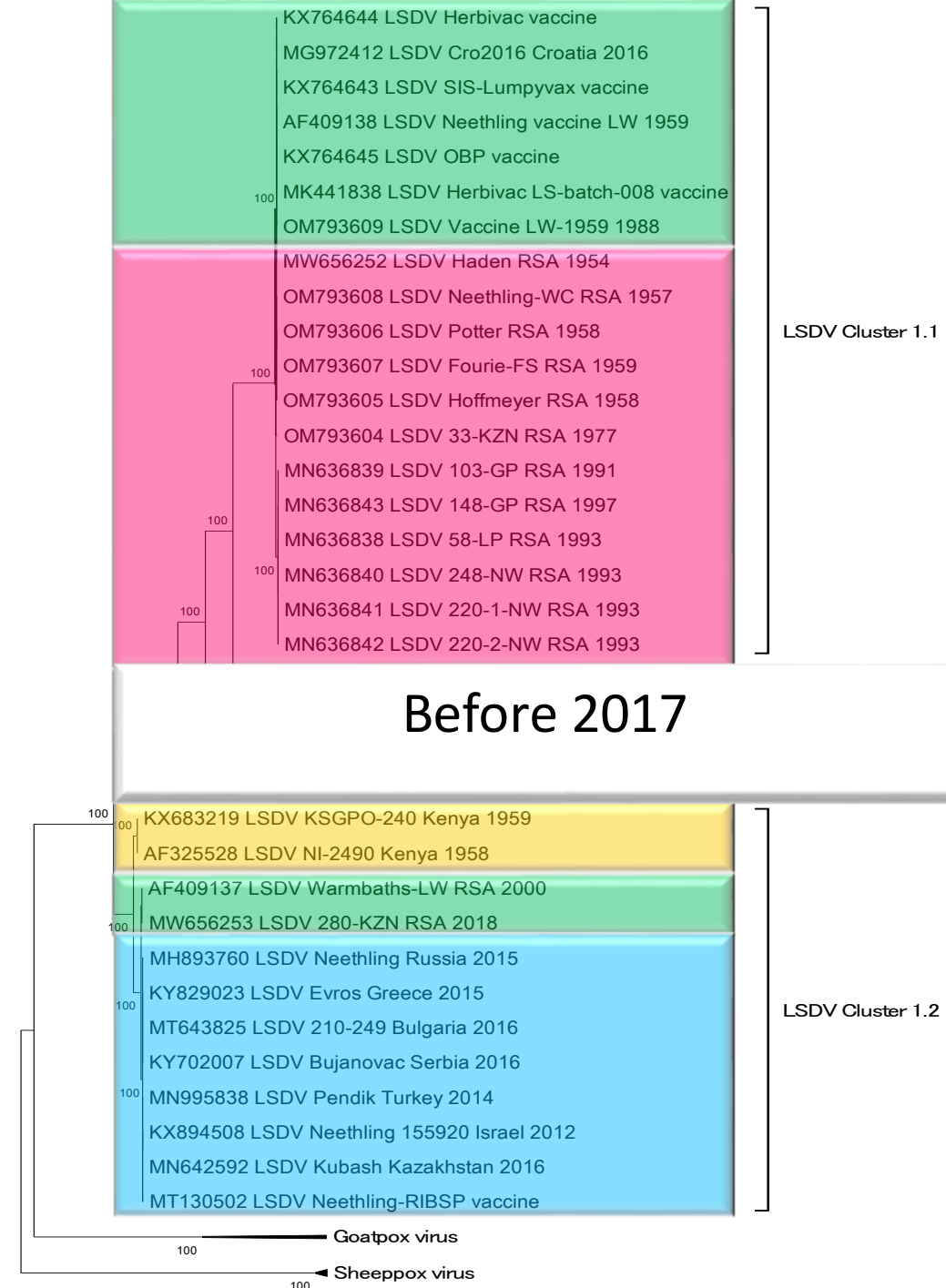
± 2,200 SNPs between cluster 1.1 and 1.2

(Kara et al., 2003; van Schalkwyk et al., 2021)

- ± 1860 SNPs in 114 ORFs
 - 57% Synonymous
 - 26% Non-Synonymous
- ± 330 (15%) IGR

Majority of the DIVA assays are based on differentiation between cluster 1.1 and 1.2. Thus not suitable to detect the field isolates of cluster 1.1

TMRCA: ± 500 years



Lumpy skin disease: Phylogenetics (Cluster 1.1)

Oldest isolate: Haden / 1954

(van Schalkwyk et al., 2021)

Prototype: Neethling-WC / 1957

(Alexander et al., 1957; van Schalkwyk et al., 2022)

- Vaccine: Neethling-LW1959 (van Rooyen et al., 1959; Kara et al., 2003)
- Vaccine: OBP, Herbivac, SIS-Lumpyvax (Mathijs et al., 2016; Douglass et al., 2019)

Wild type field isolates: South Africa 1950's, 1970's and 1990's
(van Schalkwyk et al., 2020 and 2022)

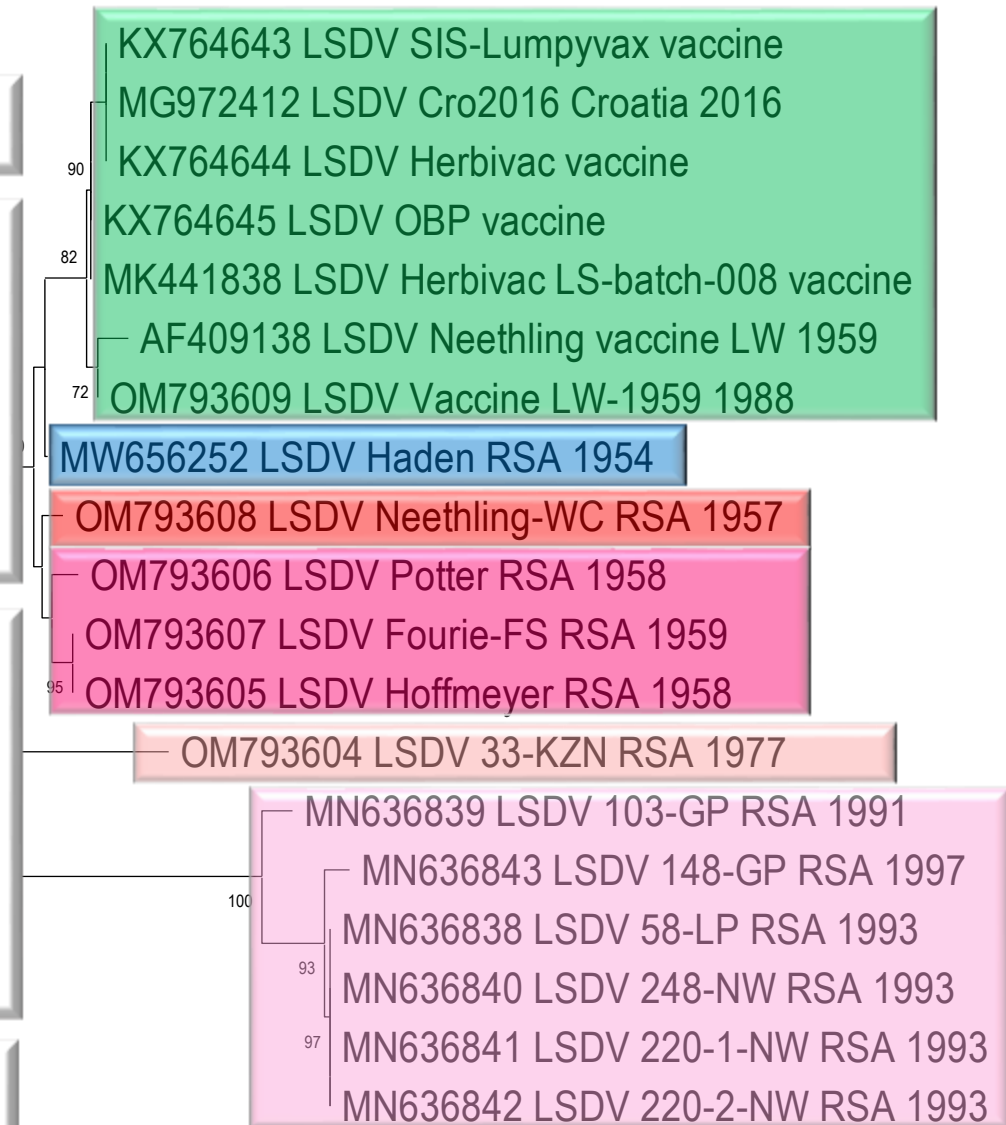
Cluster 1.1 - Attenuation

Seven SNPs between Neethling-WC/1957 and Neethling-LW1959 vaccine

(van Schalkwyk et al., 2022)

- Non-synonymous:
 - LW028 T135A
 - LW083: K663N
 - LW098: G553S
 - LW098: I625T

- Reading frame:
 - LW086: Termination
 - LW131: Termination
 - LW134: Termination



Lumpy skin disease: Phylogenetics (Cluster 1.2)

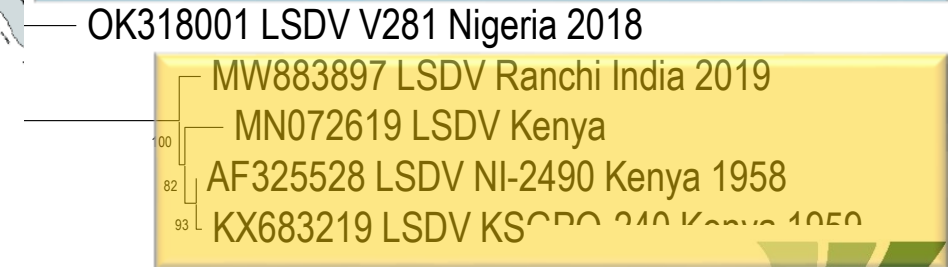
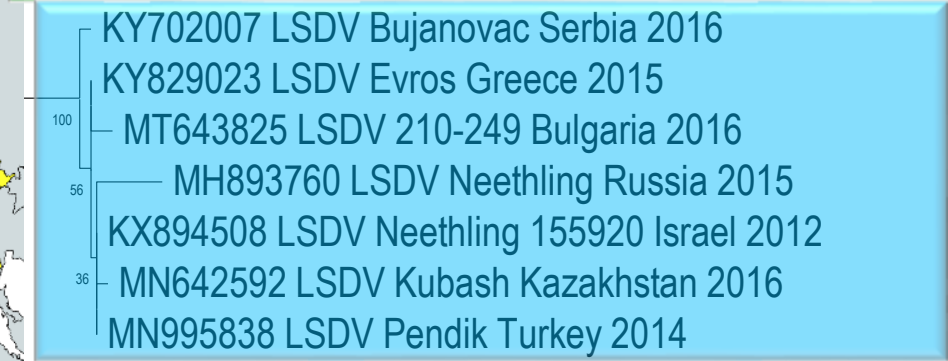
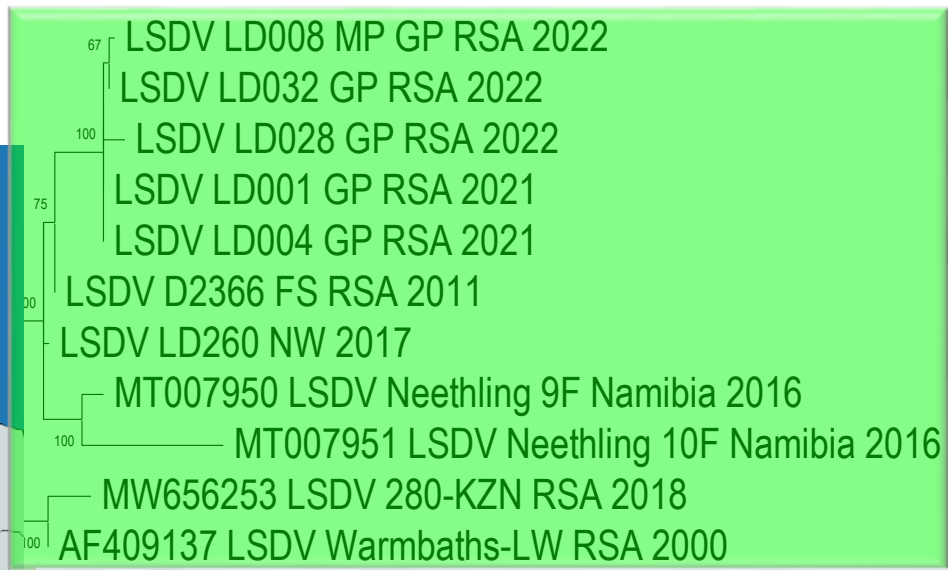
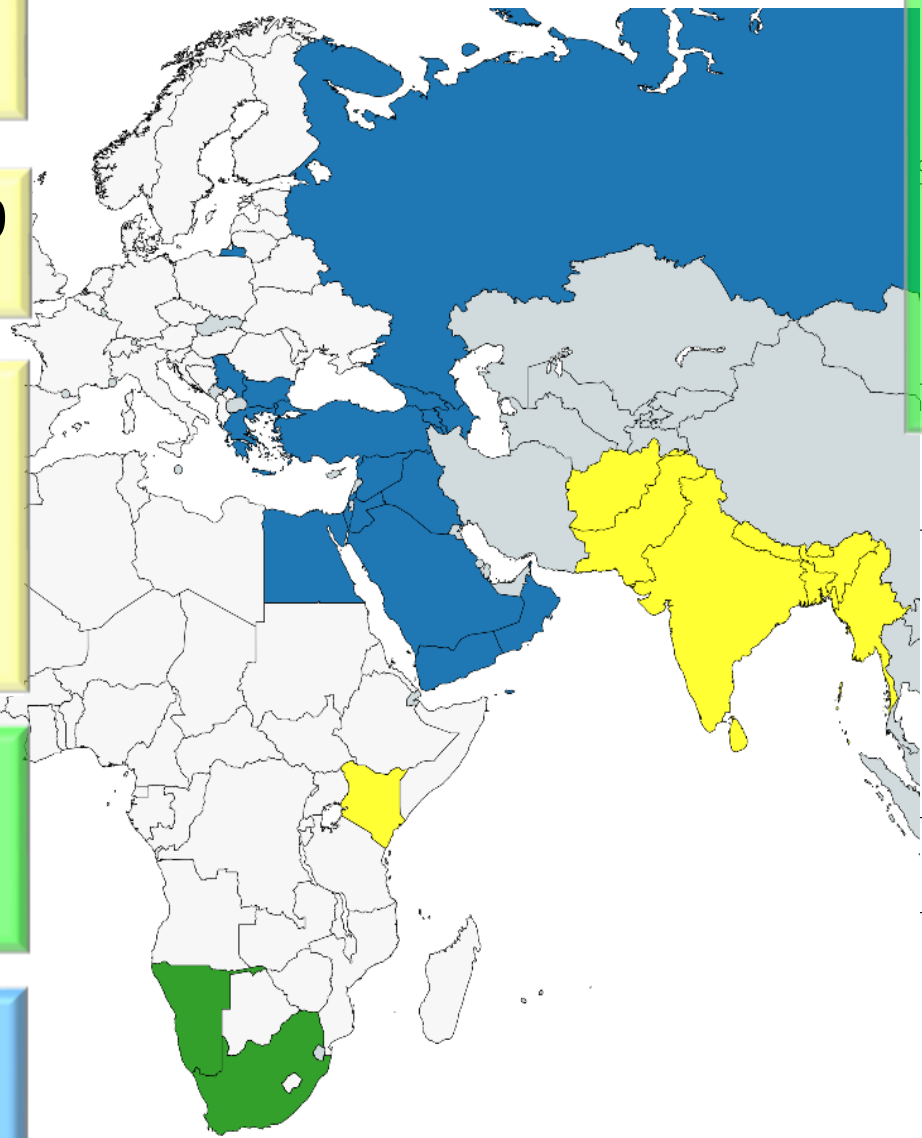
Isolate: 2490/Kenya/1958
(Tulman et al., 2000)

Vaccine: KSGPO-240/1959
(Vandenbussche et al., 2016)

Vaccine: KSGPO-240/1959. Outbreak in Bangladesh, India, Nepal and Pakistan

Isolates from southern Africa 2000 – 2022
(Kara et al., 2003)

Isolates from Middle East, Europe and Asia 2012 – 2016



Lumpy skin disease

Recombinants

Parental sequences
are both vaccines:
- Neethling-LW1959
- KSGPO-240

2.1: Saratov/Russia/2017

(Sprygin et al., 2018)

2.2: Udmurtya/Russia/2018

(Sprygin et al., 2018)

2.3: Kostanay/Kazakhstan/2018

2.4: Tyumen/Russia/2019

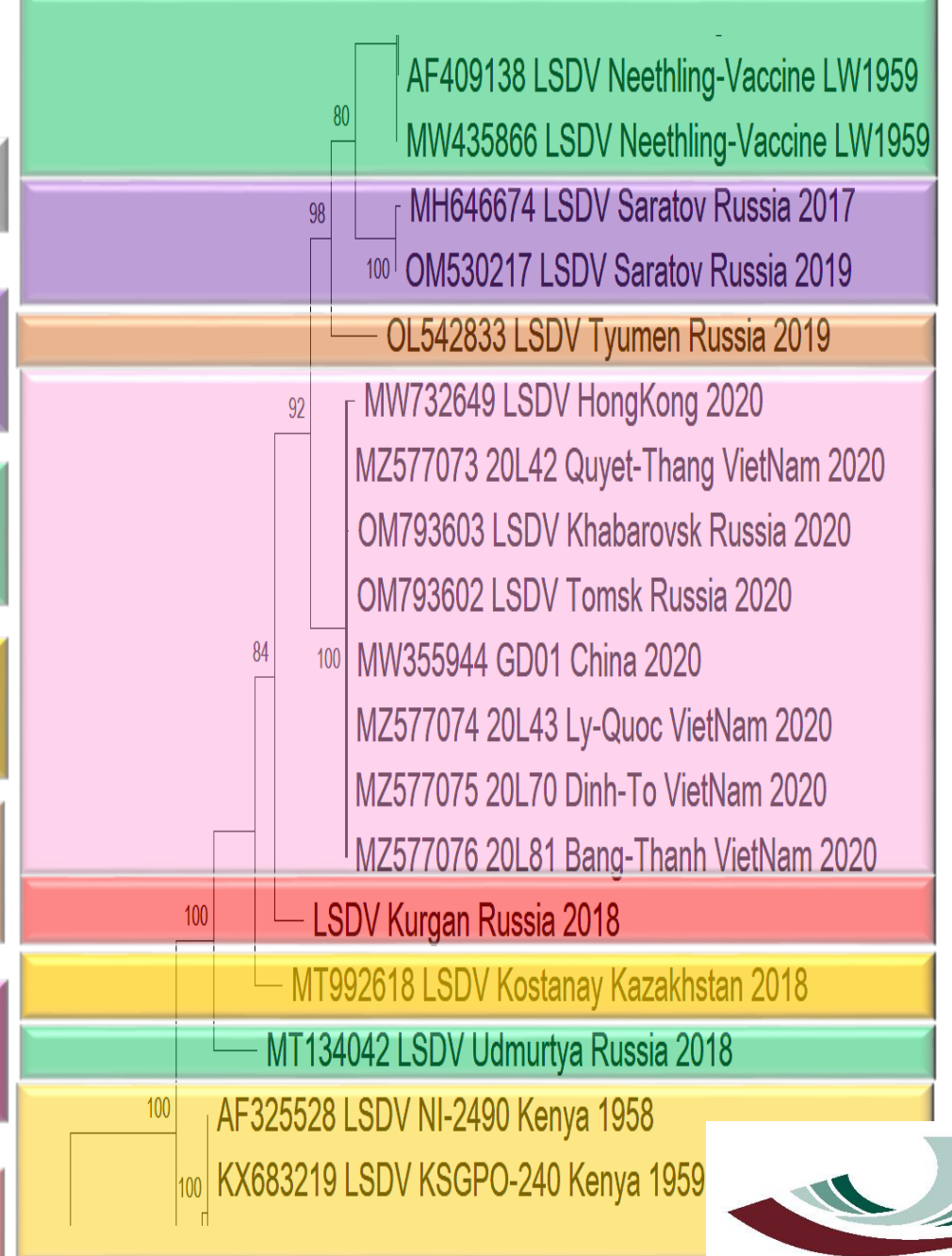
(Krotova et al., 2022)

2.5: GD01/China/2019









(Ma et al., 2021)

2.6: Kurgan/Russia/2018

(Sprygin et al., 2024)



The Importance of Quality Control of LSDV Live Attenuated Vaccines for Its Safe Application in the Field

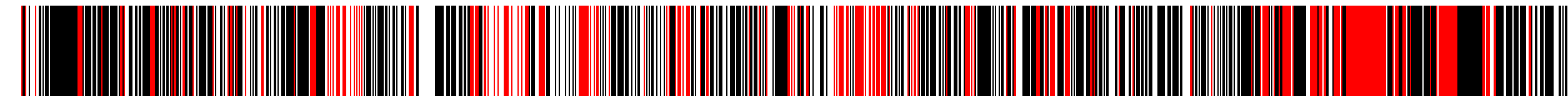
by [Andy Haegeman](#) ^{1,*}  , [Ilse De Leeuw](#) ¹ , [Meruyert Saduakassova](#) ² ,
[Willem Van Campe](#) ³ , [Laetitia Aerts](#) ⁴ , [Wannes Philips](#) ⁴ , [Akhmetzhan Sultanov](#) ² ,
[Laurent Mostin](#) ³  and [Kris De Clercq](#) ¹ 

Recombinant LSDV Strains in Asia: Vaccine Spillover or Natural Emergence?

by [Frank Vandenbussche](#) ^{1,†} , [Elisabeth Mathijs](#) ^{1,†} , [Wannes Philips](#) ¹ ,
[Meruyert Saduakassova](#) ² , [Ilse De Leeuw](#) ³ , [Akhmetzhan Sultanov](#) ² ,
[Andy Haegeman](#) ³  and [Kris De Clercq](#) ^{3,*} 

Contaminated LSD vaccine = Neethling, KSPG vaccine and GTPV

Saratov / 2017



Tuymen / 2019



China / 2019



Kurgan / 2018



Kazakhstan / 2018



Udmurtia / 2018



Non-vector-borne transmission of lumpy skin disease virus

Kononov Aleksandr¹, Byadovskaya Olga¹, Wallace B. David^{2,3}, Prutnikov Pavel¹, Pestova Yana¹, Kononova Svetlana¹, Nesterov Alexander¹, Rusaleev Vladimir¹, Lozovoy Dmitriy¹ & Sprygin Alexander¹✉

A lumpy skin disease virus which underwent a recombination event demonstrates more aggressive growth in primary cells and cattle than the classical field isolate

Svetlana Kononova¹ | Aleksandr Kononov¹ | Irina Shumilova¹ | Olga Byadovskaya¹ | Alexander Nesterov¹ | Pavel Prutnikov¹ | Shawn Babiuk² | Alexander Sprygin¹ ●

Experimentally controlled study indicates that the naturally occurring recombinant vaccine-like lumpy skin disease strain Udmurtiya/2019, detected during freezing winter in northern latitudes, is transmitted *via* indirect contact

Alexander Nesterov¹, Ali Mazloum¹, Olga Byadovskaya¹, Irina Shumilova¹, Antoinette Van Schalkwyk^{2,3}, Alena Krotova¹, Vladimir Kirpichenko⁴, Irina Donnik⁵, Ilya Chvala¹ and Alexander Sprygin^{1*}

Overwintering of recombinant lumpy skin disease virus in northern latitudes, Russia

Irina Shumilova¹ | Alena Krotova¹ | Alexander Nesterov¹ | Olga Byadovskaya¹ | Antoinette van Schalkwyk² | Alexander Sprygin¹ ●

Recombinants: Novel phenotypes

- Transmission
- Overwintering
- Aggressive growth



Lumpy skin disease: Phylogenetics

Phylogenetic analysis shows the majority of LSDV strains group into two monophyletic clusters (cluster 1.1 and 1.2) (Biswas et al., 2020; Van Schalkwyk et al., 2021).

Cluster 1.1:
Neethling Prototype
strain and vaccine

Cluster 1.1 consists of LSDV Neethling vaccine strains that are based on the LSDV/Neethling/WC-1957 type-strain (Kara et al., 2003; Van Rooyen et al., 1959; van Schalkwyk et al., 2020) and historic wild-type strains from South Africa.

Cluster 1.2:
KSGP and “wild
type”

Cluster 1.2 consists of wild-type strains from southern Africa, Kenya, the northern hemisphere, and the Kenyan KSGP O-240 commercial vaccine.

Cluster 2.1 -2.6:
Six unique
recombinant strains

In addition to these two clusters, there have recently been recombinant LSDV strains isolated from clinical cases of LSD in the field in Russia and central Asia (Flannery *et al.*, 2021; Sprygin *et al.*, 2018; 2020; Wang *et al.*, 2021). These recombinant viruses show unique patterns of accessory gene alleles, consisting of sections of both wild-type and “vaccine” LSDV strains.

- Laboratory confirmation:
 - “Gold standard” serological test: serum/virus neutralization test (SNT/VNT).
Not all animals either naturally infected or vaccinated develop LSDV neutralizing antibodies.
 - Enzyme-linked immunosorbent assay (ELISA) by IDVet (France).
 - Conventional and Real-Time Polymerase chain reaction (PCR) assays.
 - Virus isolation on cell culture (Skin nodules).

Lumpy skin disease: WOAAH Manual: B. DIAGNOSTIC TECHNIQUES

Key:
 +++ = recommended for this purpose
 ++ recommended but has limitations
 + = suitable in very limited circumstances
 – = not appropriate for this purpose

Method	Purpose					
	Population freedom from infection	Individual animal freedom from infection prior to movement	Contribute to eradication policies	Confirmation of clinical cases	Prevalence of infection – surveillance	Immune status in individual animals or populations post-vaccination
Detection of the agent						
Virus isolation	+	++	+	+++	+	–
PCR	++	+++	++	+++	+	–
Transmission electron microscopy	–	–	–	+	–	–
Detection of immune response						
VNT	++	++	++	++	++	++
IFAT	+	+	+	+	+	+
ELISA	++	++	++	++	++	++

PCR = polymerase chain reaction; VNT = virus neutralisation test;

IFAT = indirect fluorescent antibody test; ELISA = enzyme linked immunosorbent assay

Lumpy skin disease: Molecular test to differentiate vaccine and wild-type LSDV

DIVA: Differentiation of Infected from Vaccinated Animals

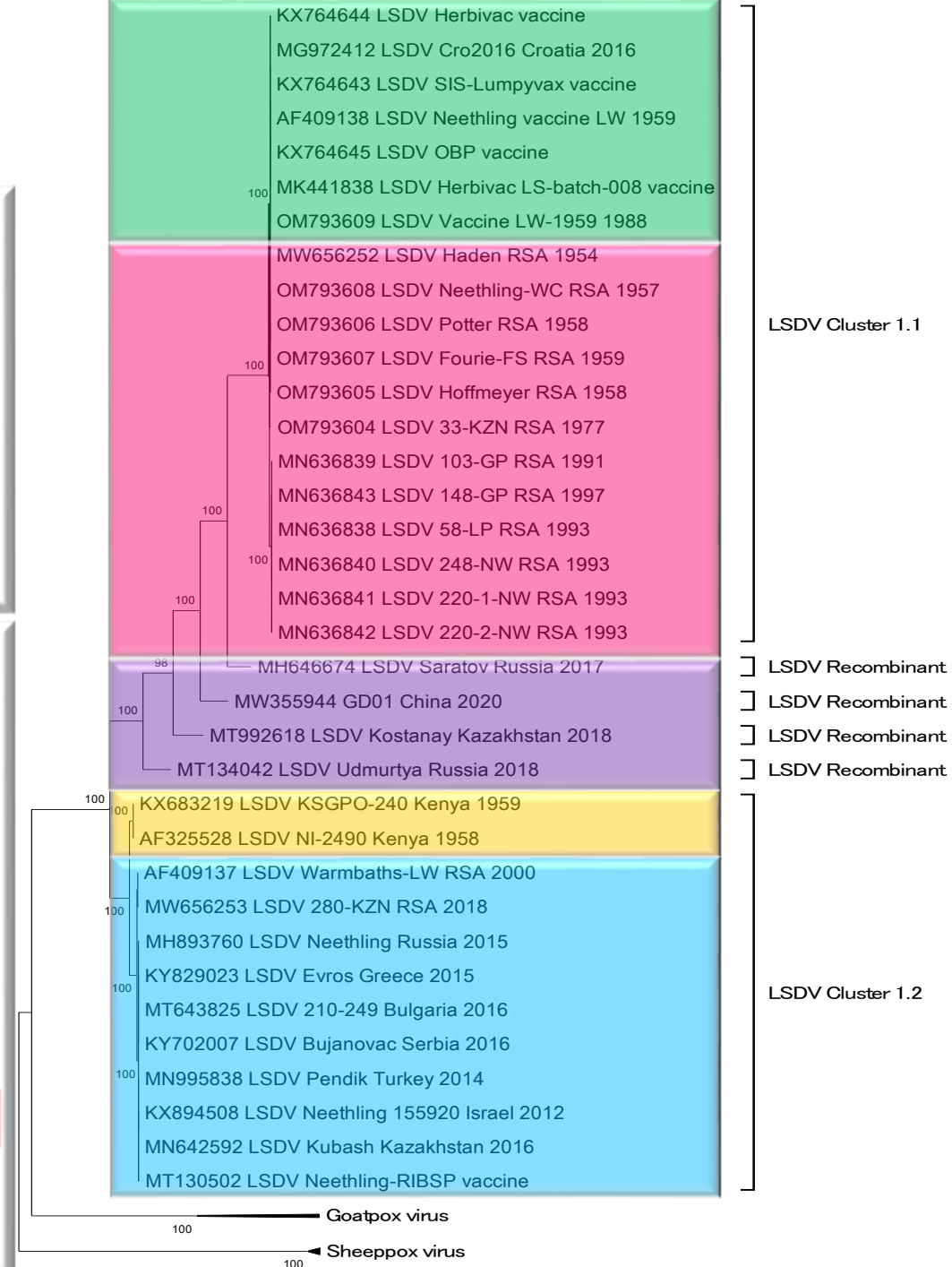
CAN differentiate between cluster 1.1 and 1.2.

CAN NOT differentiate between vaccine and wild type isolates within Cluster 1.1

CAN NOT differentiate between vaccines and novel recombinant strains (Cluster 2.1 – 2.5)

Quantitative real-time PCR assays have been designed to differentiate the “Neethling response” caused by vaccination with a **Neethling-based LSDV strains** and **wild-type LSDV strains from cluster 1.2** (Agianniotaki et al., 2017; Pestova et al., 2018; Vidanovic et al., 2016).

However, they cannot distinguish between a **LSDV Neethling vaccine strain** and the novel **recombinant LSDV strains** recently isolated from disease outbreaks in Asia (Byadovskaya et al., 2021; Flannery et al., 2021). These DIVA assays are also not capable of discriminating between LSDV Neethling vaccine strains and recently characterised **(historic) wild-type viruses from South Africa** belonging within cluster 1.1 (Van Schalkwyk et al., 2020; 2021).



Lumpy skin disease: New Markers

Kumar et al., 2023

HRM-based gap-qRT-PCR: 801bp in terminal repeat region (ITR)

- Vaccine: (Lumpi-ProVac^{Ind}) vs. Wild type: (LSDV/2019/India/Ranchi)

Haegeman et al.,
2023

Duplex qRT-PCR: LW133 and LW144

- Vaccine (Neethling) vs. Wild type: Cluster 1.2 vs. Recombinant (Cluster 2.5)

Krotova et al., 2023

PCR and Sanger sequencing: 705bp in ORF LW134

- Vaccines (Neethling and KSGPO) vs. Wild type Cluster 1.2 vs. Recombinant (Cluster 2.1, 2.2, 2.3, 2.4 and 2.5)

Lumpy skin disease: New Diagnostic tests

Nan et al., 2023

Triplex real-time PCR:

- LSDV vs GTPV vs SPPV

Liao et al., 2023

CRISPR-Cas12a:

- LSDV vs GTPV vs SPPV

Nandi et al., 2023

Isothermal PCR: 27bp in ORF LW126

- Vaccines (GTPV) vs. Wild type Cluster 1.2-KSGPO

Abdalhamed et al.,
2022

Gold nanoparticle – lateral flow test

Sthitmatee et al.,
2023

In-house ELISA using whole virus (LSD/THA/CMU/21/05)

NO DIVA

Homologous vaccines:

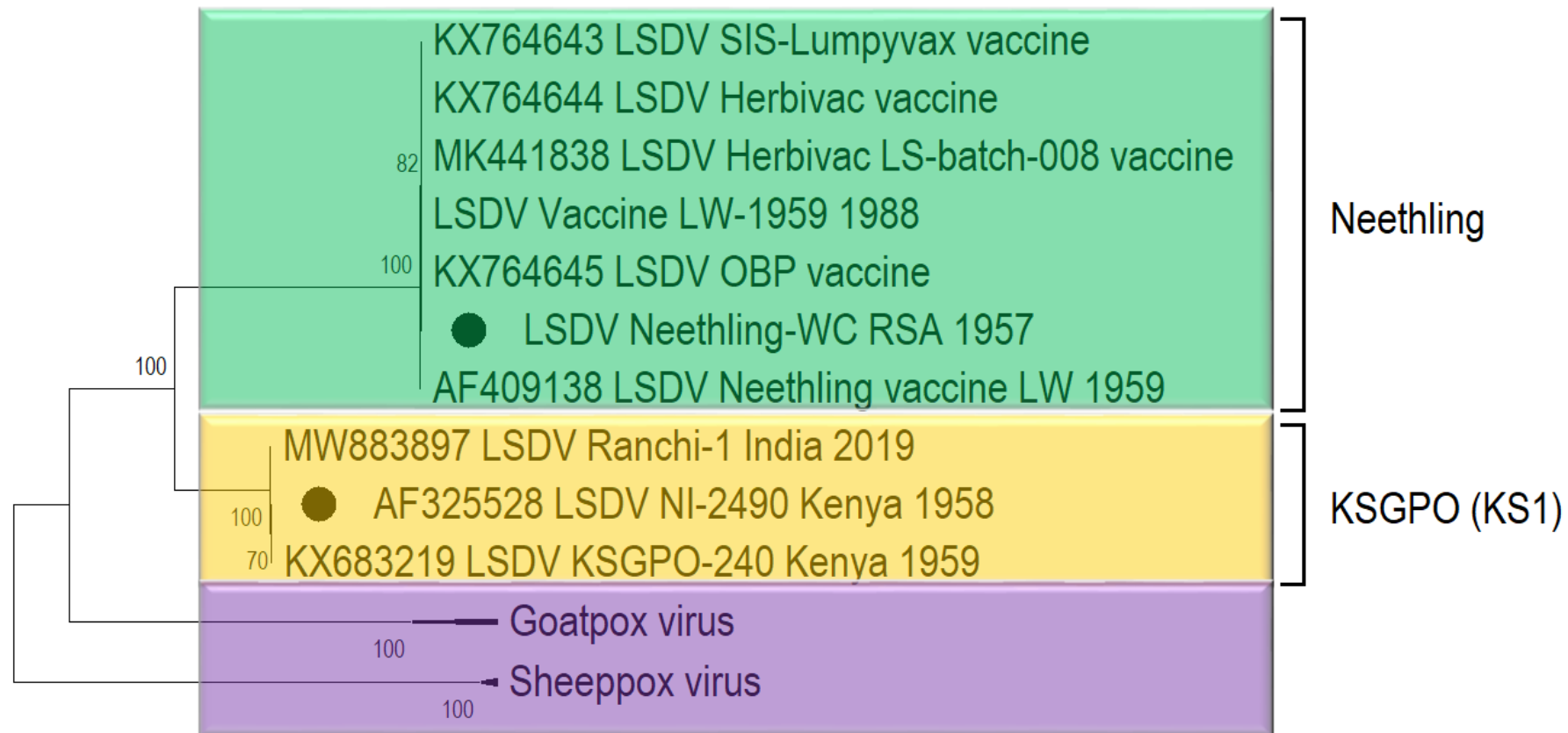
Attenuated LSDV
(Neethling and KSGPO)

Heterologous vaccines:

Goatpox (Gorgan /
Uttarkashi)
Sheeppox (NISHKI)

Haegeman et al., 2023;
Hamdi et al., 2020;
Wolf et al., 2022

DIVA



Homologous inactivated vaccines: Safe, regular boosters; <1 year immunity

New vaccines: vector-, subunit, mRNA vaccines

Lumpy skin disease: Available vaccines

Manufacturer	Product Name and Virus Strain	Target Species	Titre, Dose, Administration
Onderstepoort Biological Products (OBP) South Africa	Lumpy Skin Disease (LSD Neethling strain)	Vaccine for Cattle	$10^{3.5}$ TCID ₅₀ /dose 2 ml SC
Intervet (Pty) South Africa/MSD Animal Health	Lumpyvax™ (LSD SIS Neethling strain)	Vaccine for Cattle	$10^{4.0}$ TCID ₅₀ /dose 2 ml SC
MCI Santé Animale Morocco	Bovivax-LSD™ (LSD Kenya strain)	Vaccine for Cattle	$10^{3.5}$ TCID ₅₀ /dose 2 ml SC
Jordan Bio-Industries Center (JOVAC) Jordan	LumpyShield-N™ (LSD Neethling strain)	Vaccine for Cattle	$10^{4.0}$ TCID ₅₀ /dose 2 ml SC
Jordan Bio-Industries Center (JOVAC) Jordan	Caprivac™ (Gorgan GTP strain)	Vaccine for Cattle	
Middle East for Vaccines (MEVAC) Egypt	MEVAC LSD (LSD Neethling strain)	Vaccine for Cattle	$10^{3.5}$ TCID ₅₀ /dose 2 ml SC
National Veterinary Institute (NVI) Ethiopia	Lumpy Skin Disease vaccine (LSD Neethling strain)	Vaccine for Cattle	$10^{3.0}$ TCID ₅₀ /dose 2 ml SC
Kenya Veterinary Vaccines Production Institute (KEVEVAPI)	Lumpivax™ (LSD Neethling strain)	Vaccine for Cattle	Not known TCID ₅₀ /dose 2 ml SC
Pendik Veterinary Control Institute/ Ministry of Agriculture, Turkey	Penpox-M™ Live SPPV (Bakirköy SPPV strain)	Vaccine for Cattle	$10^{2.5}$ TCID ₅₀ /dose 2 ml SC
Vetal Company Turkey	Poxvac™ (Bakirköy SPPV strain)	Vaccine for Sheep and Cattle	$10^{2.5}$ TCID ₅₀ /dose 2 ml SC
	Lumpyvac™ (LSD Neethling strain)	Vaccine for Cattle	$10^{3.5}$ TCID ₅₀ /dose 2 ml SC
Dollvet Turkey	Poxdoll™ (Bakirköy SPPV strain)	Vaccine for Sheep, goats and Cattle	$10^{2.5}$ TCID ₅₀ /dose 2 ml SC
	LSD-NDOLL (LSD Neethling strain)	Vaccine for Cattle	$10^{3.5}$ TCID ₅₀ /dose 2 ml SC
FGBI-Federal Centre for Animal Health Russia	Sheep Pox Cultyral Dry™ (Arriah (NISHKI) SPPV strain)	Vaccine for Sheep and Cattle	Not known TCID ₅₀ /dose
ABIC, Israel	RM 65 Sheeppox (Yugoslavia RM65)	Vaccine for Sheep and Cattle	

Tuppurainen et al., 2021

Lumpy skin disease: Vaccine testing (literature study by Pravesh Kara)

Breed Age Gender	Construct / Vaccine Dose Route & Volume	Number of animals	Fever (%)	Start of fever (Ave Days)	Inoculation site reaction (%)	>1 nodule (%)	Clinical reaction [other than fever] (%)	Shedding (PCR) (%)	Nodule (PCR) (%)	Viremia (PCR / VI) (%)	VNT 1 (%)	VNT 2 (%)	ELISA (%)	IPMA (%)	IFIT / IFAT (%)	Reference
Holstein - Friesen 6 months Male	LSD Neethling OBP [1.4x10 ⁷ TCID ₅₀] SC Route: 2ml	5	0/5 (0%)	N/A	1/5 (20%)	0/5 (0%)	1/5 (20%)	Nd	0/1 (0%)	PCR 0/5 VI 0/5	0/5 (0%)	nd	nd	nd	nd	Kara et al., 2018
Holstein 6 months Male	LSD Neethling OBP [1x10 ^{3.5} TCID ₅₀] SC Route: 2ml	7	6/7 (86%)	1 dpv (4.2 days)	0/7 (0%)	0/7 (0%)	0/7 (0%)	Nd	0/7 (0%)	0/7 (0%)	1/7 (14%)	3/7 (43%)	nd	7/7 (100%)	nd	Haegeman et al., 2021a
	LSD Neethling Lumpyvax [1x10 ⁴ TCID ₅₀] SC Route	7	7/7 (100%)	1 dpv (10.3 days)	0/7 (0%)	0/7 (0%)	0/7 (0%)	Nd	0/7 (0%)	0/7 (0%)	1/7 (14%)	4/7 (57%)	nd	5/7 (72%)	nd	
	LSD Neethling HerbivacLS [1x10 ^{2.5} TCID ₅₀] SC Route	7	4/7 (57%)	1 dpv (3 days)	0/7 (0%)	3/7 (43%)	3/7 (43%)	nd	3/7 (43%)	4/7 (57%)	3/7 (43%)	5/7 (71%)	nd	6/7 (86%)	nd	
	LSD Neethling (O variant) MCI [1x10 ³ TCID ₅₀] SC Route	7	4/7 (57%)	1 dpv (3.8 days)	3/7 (43%)	2/7 (29%)	3/7 (43%)	nd	2/7 (29%)	2/7 (29%)	4/7 (57%)	3/7 (43%)	nd	7/7 (100%)	nd	
	LSD KSGP Kenyavac [1x10 ^{2.5} TCID ₅₀] SC Route	7	5/7 (71%)	1 dpv (6.8 days)	0/7 (0%)	0/7 (0%)	0/7 (0%)	nd	0/7 (0%)	0/7 (0%)	0/7 (0%)	2/7 (29%)	nd	5/7 (72%)	nd	
Holstein-cross 4-6 months	LSD Neethling (LSD _{NT}) [1x10 ⁴ TCID ₅₀] SC Route: 2ml	15	7/15 (47%)	2 dpv (2 days)	0/15 (0%)	1/15 (7%)	1/15 (7%)	1/15 (7%)	1/15 (7%)	nd	7/15 (47%)	nd	nd	nd	nd	Bamouh et al., 2021
	LSD Neethling (LSD _{NT}) [1x10 ⁵ TCID ₅₀] SC Route: 2ml	30	13/30 (43%)	NI (4.3 days)	2/30 (7%)	2/30 (7%)	2/30 (7%)	2/30 (7%)	2/30 (7%)	nd	23/30 (73%)	nd	nd	nd	nd	
	LSD KSGP O-240 [1x10 ⁴ TCID ₅₀] SC Route: 2ml	12	5/12 (42%)	NI (5.6 days)	0/12 (0%)	0/12 (0%)	0/12 (0%)	0/12 (0%)	0/12 (0%)	0/12 (0%)	12/12 (100%)	nd	nd	nd	nd	
	LSD KSGP O-240 [1x10 ⁵ TCID ₅₀] SC Route: 2ml	12	9/12 (75%)	NI (3.8 days)	1/12 (8%)	3/12 (25%)	3/12 (25%)	3/12 (25%)	3/12 (25%)	3/12 (25%)	12/12 (100%)	nd	nd	nd	nd	
Morocco (Zebu) 6-8 months	LSD Neethling OBP [1x10 ⁴ TCID ₅₀] SC Route	15	1/15 (7%)	1 dpv (1 day only)	0/15 (0%)	0/15 (0%)	0/15 (0%)	0/15 (0%)	0/15 (0%)	nd	7/15 (47%)	nd	nd	nd	nd	Hamdi et al., 2020
Holstein-Friesen 9-10 months	LSD Neethling (Nt _{hd}) [1x10 ⁷ TCID ₅₀] IV Route: 3ml & SC Route: 1ml	6	0/6 (0%)	N/A	2/6 (33%)	0/6 (0%)	0/6 (0%)	2/6 (33%)	0/6 (0%)	4/6 (67%)	4/6 (67%)	nd	5/6 (83%)	nd	6/6 (100%)	Moller et al., 2019
Holstein-Friesen 4-6 months	LSD Neethling HerbivacLS [1x10 ^{2.5} TCID ₅₀] SC Route: 2ml	6	3/6 (50%)	5-6 dpv. (4-6 days)	5/6 (83%)	0/6 (0%)	5/6 (83%)	0/6 (0%)	nd	4/6 (67%)	5/6 (83%)	nd	5/6 (83%)	nd	nd	Wolff et al., 2020
Borana (Zebu) 12-24 months	LSD Neethling (NVI, Ethiopia) [1x10 ^{4.5} TCID ₅₀] SC Route	5	0/5 (0%)	N/A	0/5 (0%)	0/5 (0%)	0/5 (0%)	nd	nd	nd	nd	nd	nd	nd	0/5 (0%)	Gari et al., 2015
	LSD Neethling (NVI, Ethiopia) [1x10 ^{3.5} TCID ₅₀] SC Route	5	0/5 (0%)	N/A	0/5 (0%)	0/5 (0%)	0/5 (0%)	nd	nd	nd	nd	nd	nd	nd	0/5 (0%)	
	LSD KSGP O-180 [1x10 ^{4.5} TCID ₅₀] SC Route	5	0/5 (0%)	N/A	0/5 (0%)	0/5 (0%)	0/5 (0%)	nd	nd	nd	nd	nd	nd	nd	0/5 (0%)	
	LSD KSGP O-180 [1x10 ^{3.5} TCID ₅₀] SC Route	5	0/5 (0%)	N/A	0/5 (0%)	0/5 (0%)	0/5 (0%)	nd	nd	nd	nd	nd	nd	nd	0/5 (0%)	
Dexter 11-16 months Male	LSD Neethling OBP [1x10 ^{3.5} TCID ₅₀] SC Route: Vaccination 1	6	0/6 (0%)	N/A	0/6 (0%)	0/6 (0%)	0/6 (0%)	nd	nd	0/6 (0%)	0/6 (0%)	nd	nd	nd	nd	Osuagwuh et al., 2007
	LSD Neethling OBP [1x10 ^{3.5} TCID ₅₀] SC Route: Repeat Vaccination 21 dpv	6	0/6 (0%)	N/A	0/6 (0%)	0/6 (0%)	0/6 (0%)	nd	nd	0/6 (0%)	4/6 (67%)	nd	nd	nd	nd	
Holstein 6 months Male	LSD Lumpivax (KEVEVAPI, Kenya) [Dose: NI] SC Route: 2ml	7	7/7 (100%)	NI	7/7 (100%)	2/7 (29%)	7/7 (100%)	nd	3/3 (100%)	1/7 (14%)	nd	nd	1/7 (14%)	7/7 (100%)	nd	Haegeman et al., 2021b
Breed: NI 6-9 months Male	LSD Lumpi-ProVac Ind [1x10 ^{3.5} TCID ₅₀] Route: NI	8	3/8 (37.5%)	NI	0/8 (0%)	0/8 (0%)	Viremia 5/8 (62.5%) 3dpv only	0/8 (0%)	0/8 (0%)	PCR 5/8 (62.5%) 3dpv only	7/8 (87.5%)	nd	nd	nd	nd	Kumar et al., 2022

Not all animals naturally infected or vaccinated develop neutralizing antibodies against LSDV or they develop low levels of antibodies undetectable by current serological assays



Lumpy skin disease: South Africa

Endemic since 1945

Annual outbreaks – Summer months

Diagnostic – Laboratory confirmation

- Molecular: Realtime PCR
- Serology: VNT
- Serology: IDVet ELISA
- Complete genome sequencing

Vaccination: Live attenuate Neethling vaccine:

- OBP vaccine Onderstepoort Biological Products, South Africa
- Herbivac - Deltamune, South Africa
- Lumpyvax - MSD Animal Health, South Africa
- Annual vaccination - encourage

Education and information: 5 Languages

LUMPY SKIN DISEASE

What is lumpy skin disease?

- ✓ Poxvirus infecting cattle
- ✓ Symptoms include:

- ✓ Fever
- ✓ Skin nodules (Lumps)
- ✓ Ocular discharge
- ✓ Nasal discharge
- ✓ ↓ milk production

Lameness and Depression

Economic impact

- Decrease in milk production
- Decrease in value of hide
- Decrease in body condition
- Decrease in growth rate
- Decrease in international trade
- Sterility in bulls
- Infertility in cows

How does it spread?

- Movement of sick animals
- Mechanically via insects
- Contaminated feed
- Shared water troughs
- Direct contact
- Indirect contact

Can humans get LSD?





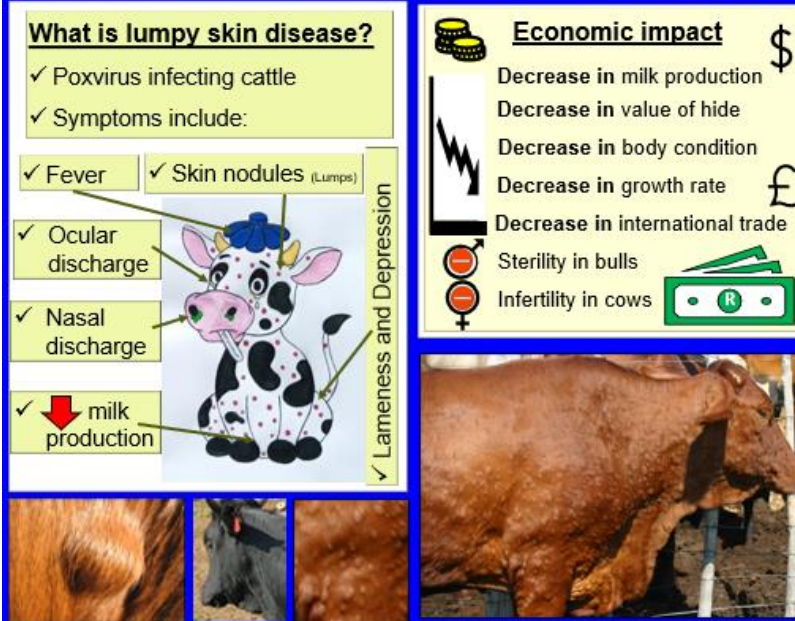
No, but cattle and wildlife such as springbok and giraffe can.

How to prevent LSD?

- ✓ Vaccinate cattle
- ✓ Vaccinate only healthy cattle
- ✓ Vaccinate annually

What should you do when you see LSD?

- ✓ Contact Veterinarian
- ✓ Contact Animal Health Technician



Questions?

Antoinette van Schalkwyk

Agricultural Research Council – Onderstepoort Veterinary Institute

vanschalkwyka1@arc.agric.za



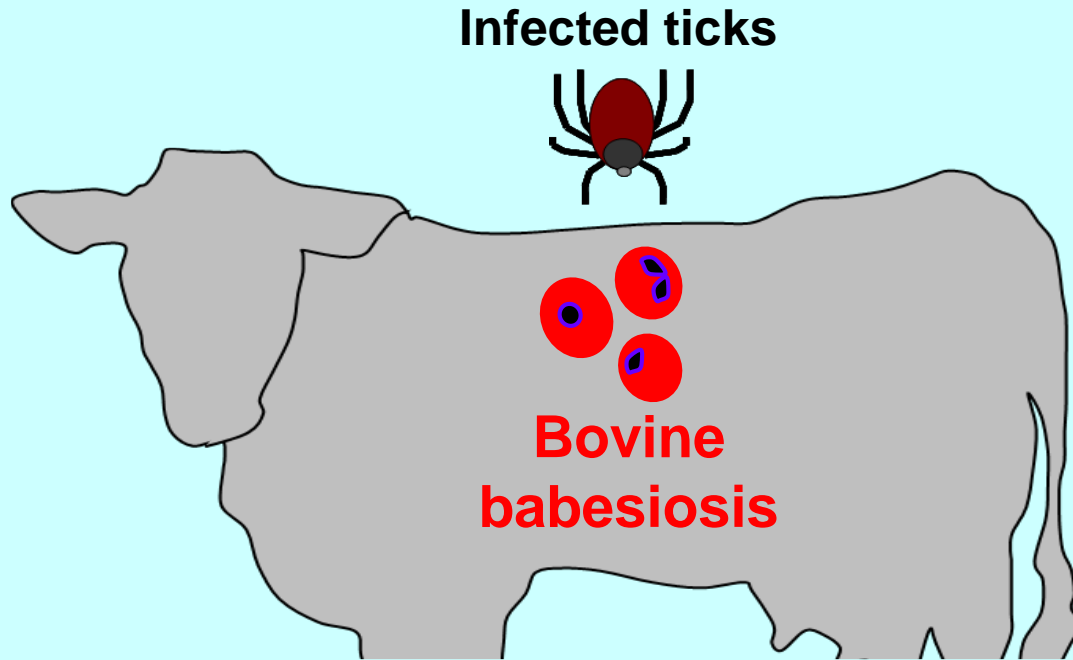
Updates on bovine babesiosis -distribution and diagnosis-

Thillaiampalam Sivakumar



**National Research Center for Protozoan Diseases
Obihiro University of Agriculture and Veterinary Medicine, Japan**

Bovine babesiosis and its economic importance

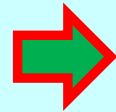


Control of bovine babesiosis is important.

Production loss

Death

Treatment



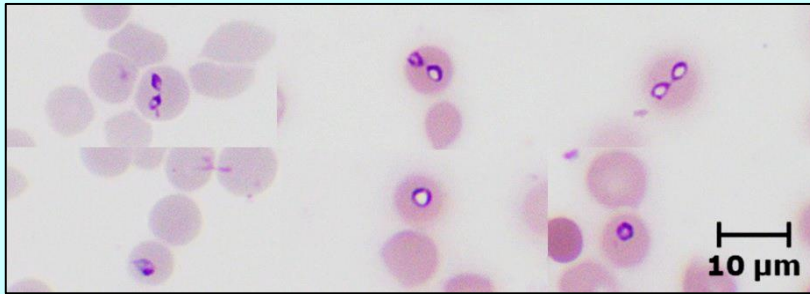
Economic loss

Tick control

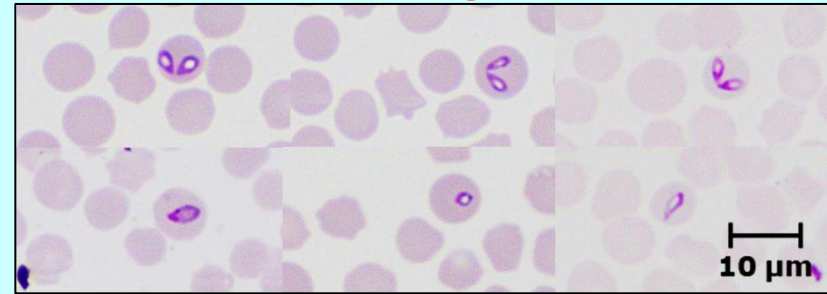
Impaired trade

Babesia species capable of causing clinical bovine babesiosis

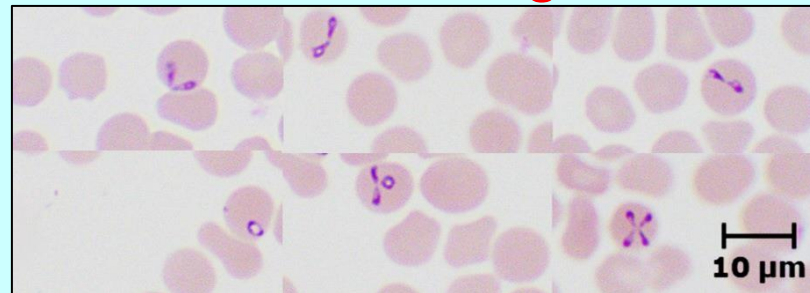
Babesia bovis



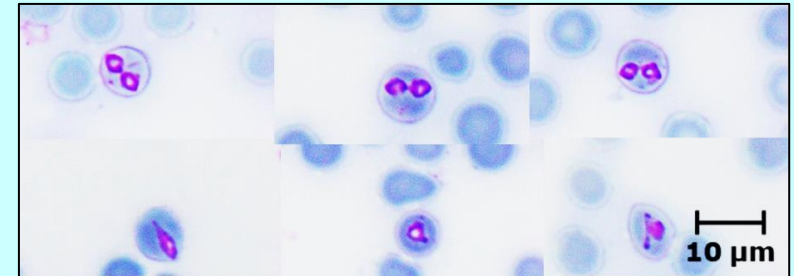
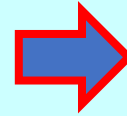
Babesia bigemina



Babesia divergens



Babesia naoakii - a new species that can cause clinical bovine babesiosis



Sivakumar et al. *Parasites & Vectors* (2022) 15:299
<https://doi.org/10.1186/s13071-022-05374-9>

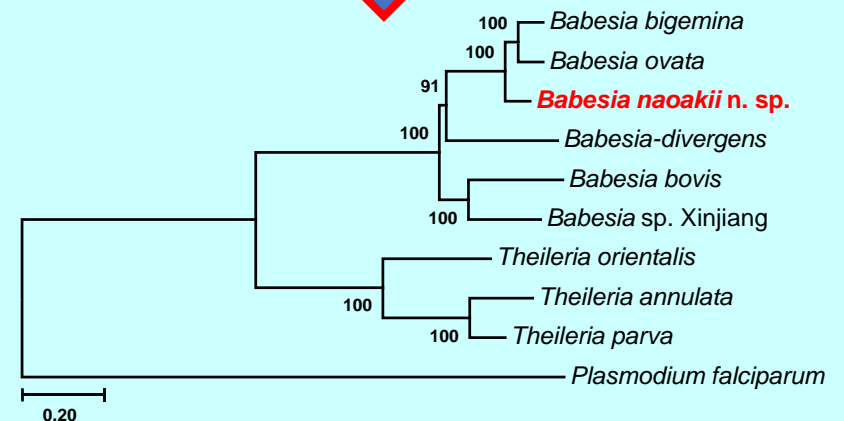
Parasites & Vectors

BRIEF REPORT

Open Access

Phylogenetic analyses of the mitochondrial, plastid, and nuclear genes of *Babesia* sp. Mymensingh and its naming as *Babesia naoakii* n. sp.

Thillaiampalam Sivakumar¹, Bumduuren Tuvshintulga¹, Davaajav Otgonsuren¹, Enkhbaatar Batmagnai¹, Believe Ahebor¹, Hemal Kothalawala², Singarayar Caniciyas Vimalakumar³, Seekkuge Susil Priyantha Silva⁴, Junya Yamagishi⁵ and Naoaki Yokoyama^{1,6*}



Tick vectors of bovine *Babesia* species

B. bovis

Rhipicephalus microplus

R. annulatus

R. geigyi

B. bigemina

R. microplus

R. decoloratus

R. annulatus

R. geigyi

R. evertsi

B. divergens

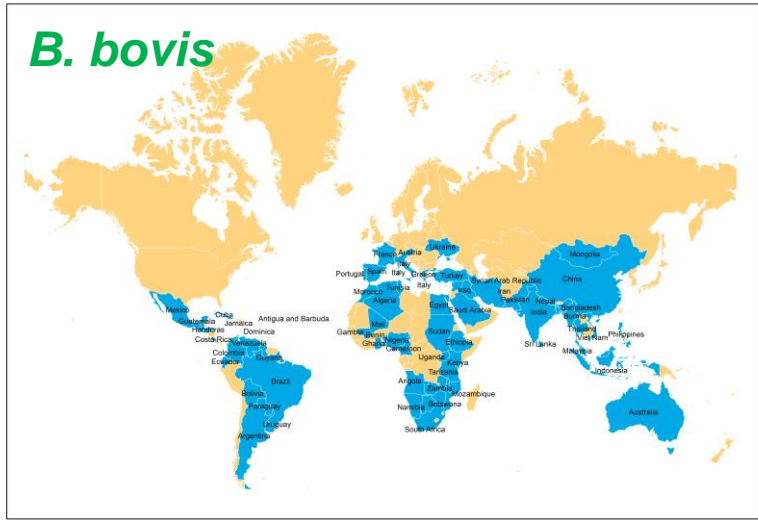
Ixodes ricinus

B. naoakii

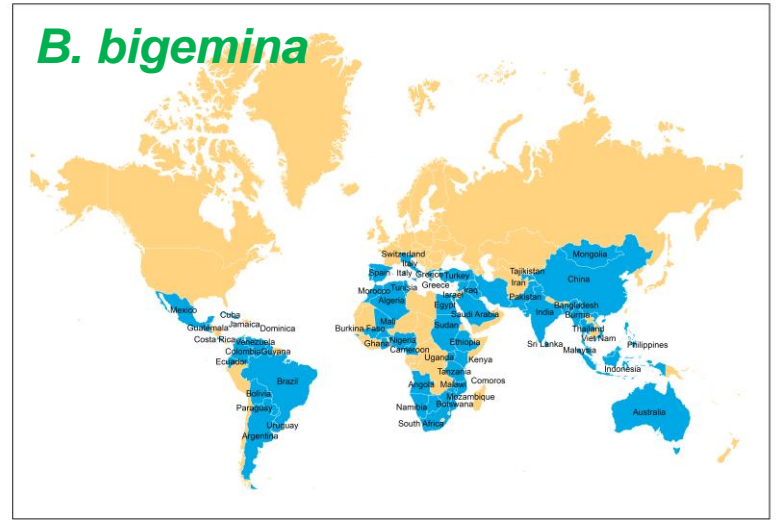
Undetermined

Global distribution of bovine *Babesia* species

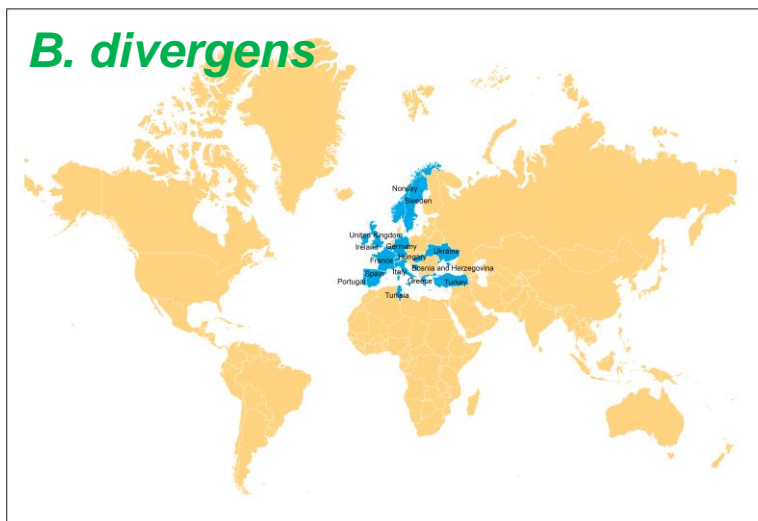
B. bovis



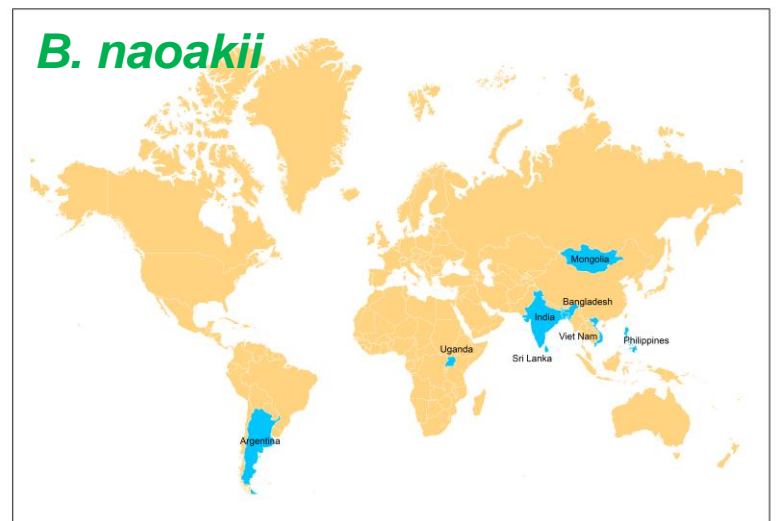
B. bigemina



B. divergens



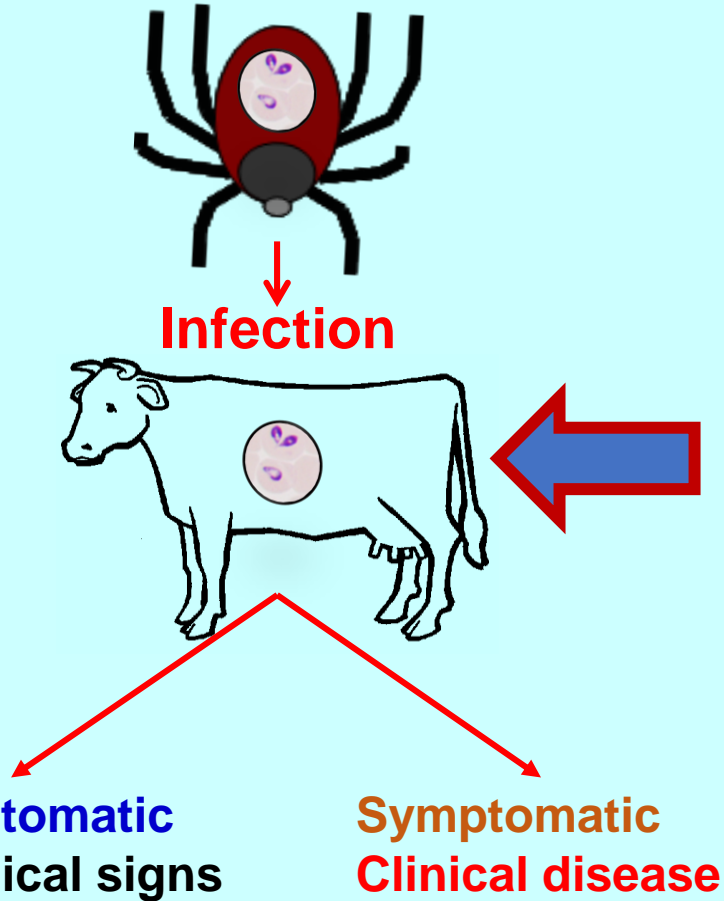
B. naoakii



Diagnosis of bovine babesiosis

1. Risk factors
2. Clinical diagnosis
3. Parasitological diagnosis
4. Molecular diagnosis

Risk factors for clinical bovine babesiosis



1. Age

Calves are resistant

2. Acquired immunity

Acquired immunity protects cattle.

3. Management practices

Cattle managed extensively are more likely to develop immunity.

4. Endemic stability

No or sporadic cases

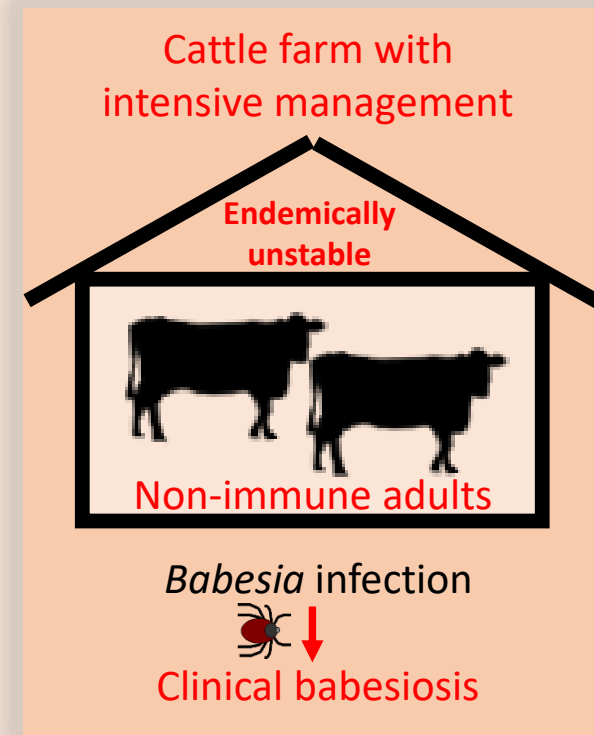
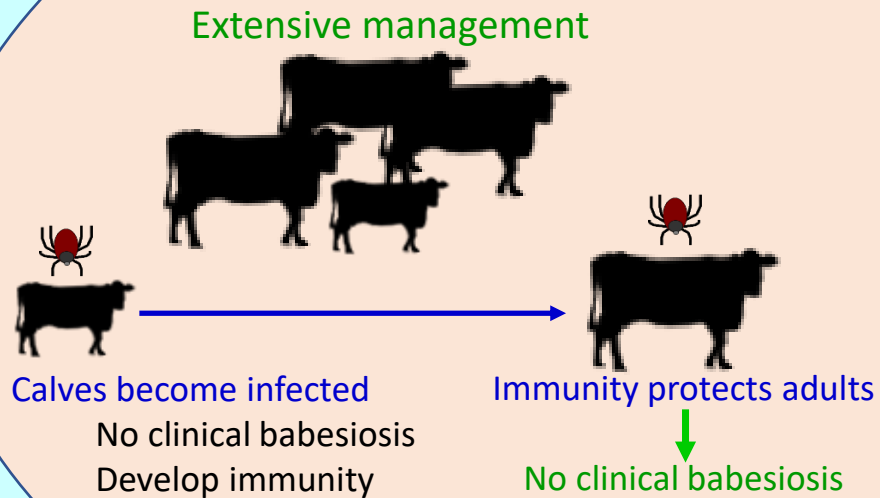
5. Cattle breed

Bos indicus is resistant, but not *Bos taurus*.

Clinical bovine babesiosis in endemic countries

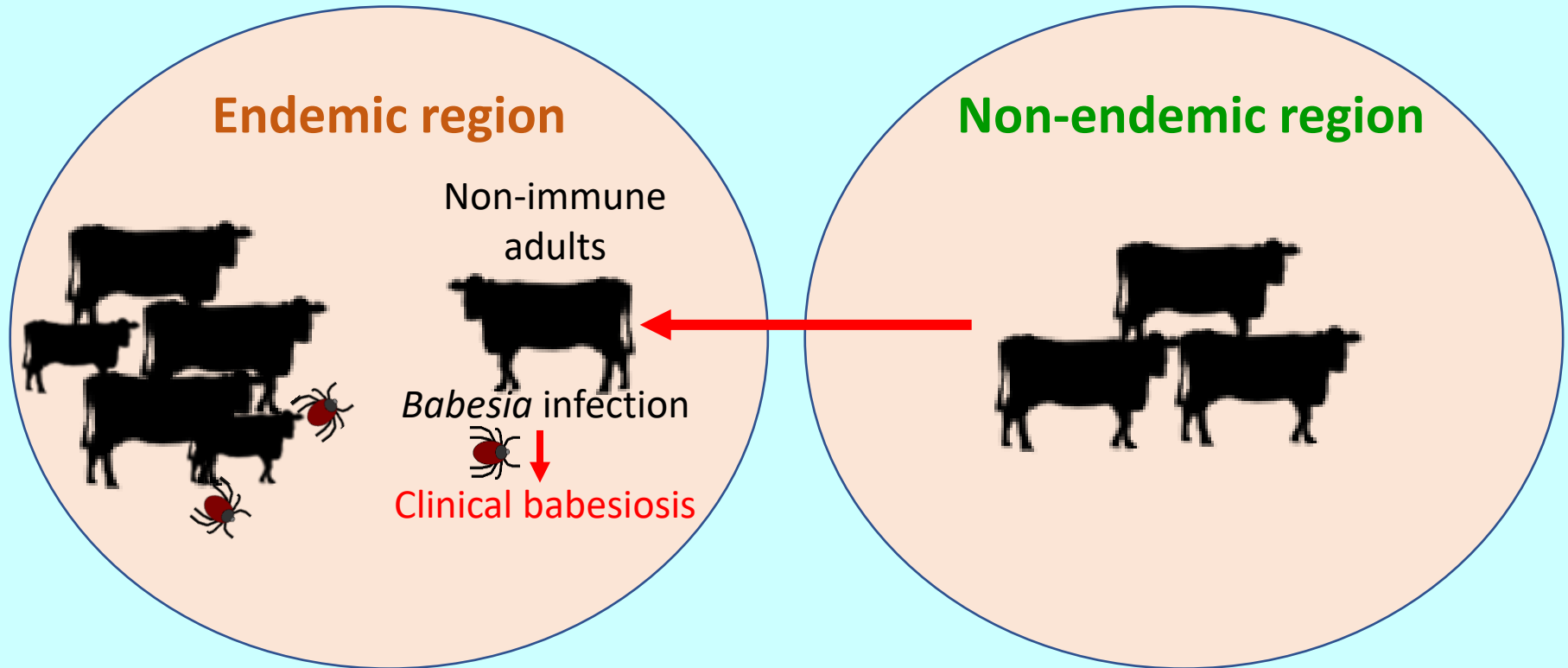
Scenario 1: endemic instability

Endemic region



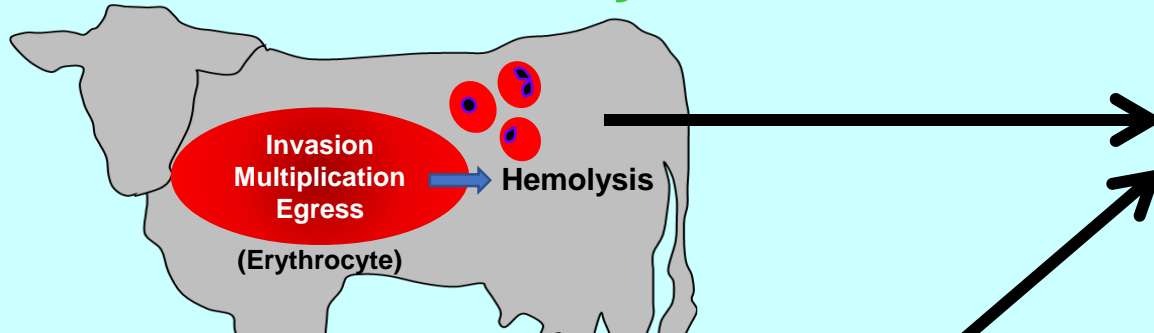
Clinical bovine babesiosis in endemic countries

Scenario 2: Introduction of naïve cattle



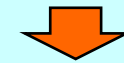
Clinical diagnosis

Intravascular hemolysis



B. bovis
B. bigemina
B. divergens
B. naoakii

High fever
Anemia
Jaundice

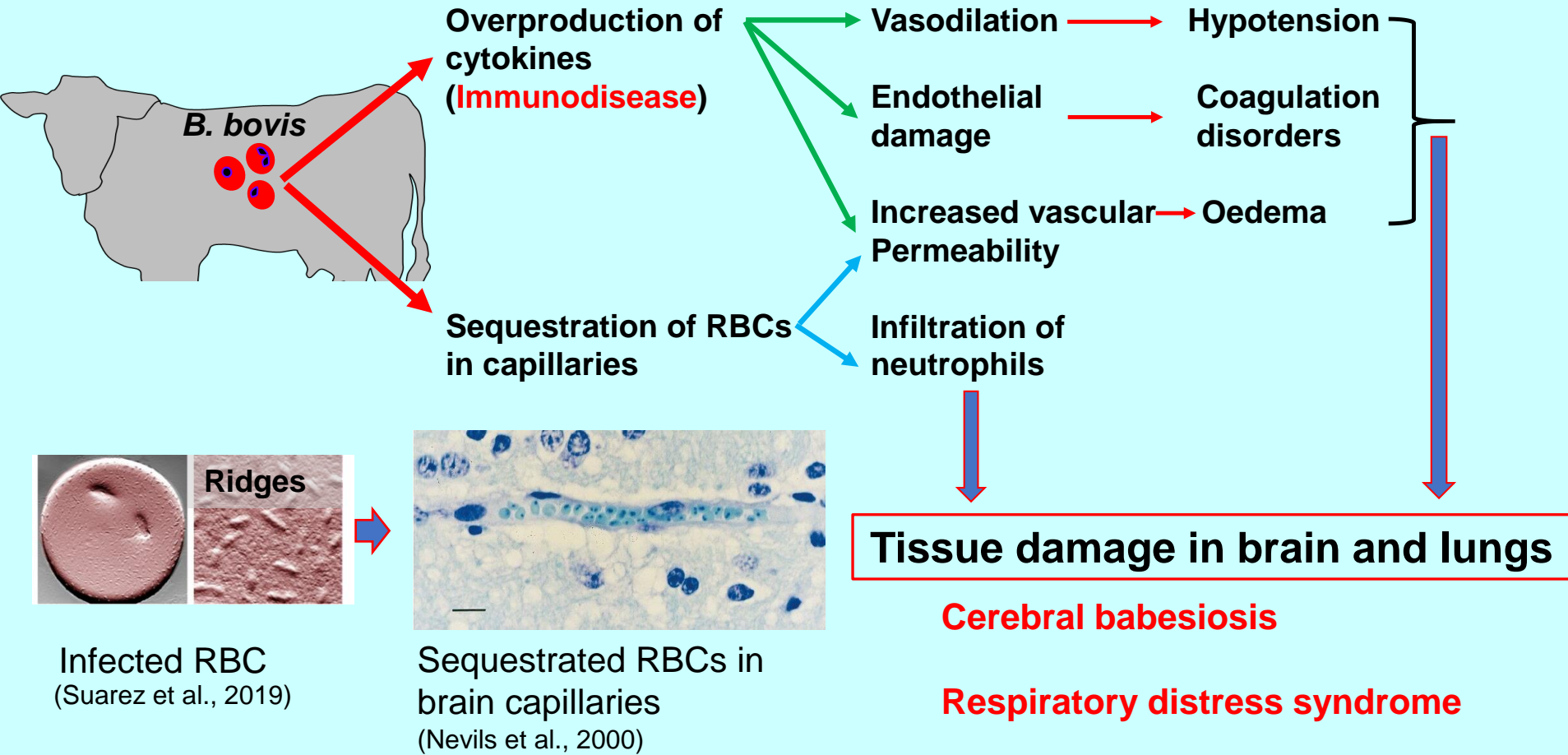


Hypoxia

Hemoglobinuria



Bovine babesiosis caused by *B. bovis*



Parasitological diagnosis

Capillary blood → Thin smears → Staining → Microscopy

Characteristics	<i>B. bovis</i>	<i>B. divergens</i>	<i>B. bigemina</i>	<i>B. naoakii</i>
Size (µm)	Small 1–1.5 × 0.5–1.0	Small 1–1.5 × 0.5–1.0	Large 3–3.5 × 1–1.5	Large 2.3–3 × 1.6–2.2
Location in RBC	Central	Rim	Entire RBC	Entire RBC
Angle between paired merozoites	Obtuse	Obtuse	Acute	Obtuse
Parasitemia in acute infection	Low (<1%)	High	High	High

Molecular diagnosis

Blood → DNA → PCR → Electrophoresis

Babesia species

Target

B. bovis

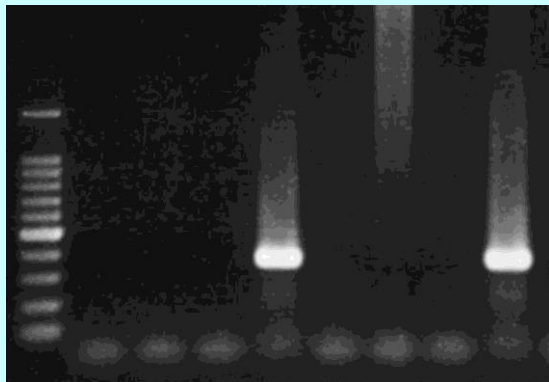
RAP-1, cytochrome b

B. bigemina

AMA-1, RAP-1, cytochrome b

B. naoakii

AMA-1



Specific and sensitive

Early diagnosis is important.

A survey of clinical bovine babesiosis in Sri Lanka

No.	Breed	Age	RBC indices			<i>Babesia</i> spp.	Treatment	Prognosis
			HGB	HCT	RBC			
1	Jersey	4	ND	ND	ND	<i>Bbo</i> + <i>Bbi</i>	Diminazene	Recovered
2	Friesian	3	8.4	22.6	5.65	<i>Bbi</i>	Diminazene	Recovered
3	Jersey	4	9.6	27.6	5.94	<i>Bbi</i>	Diminazene	Recovered
4	Friesian	4	6.8	19.6	3.94	<i>Bna</i>	Diminazene	Recovered
5	Jersey	5	2.8	7.9	1.38	<i>Bbo</i> + <i>Bbi</i>	Diminazene	Died
6	Jersey	4	8.7	24	5.59	<i>Bbo</i> + <i>Bbi</i>	Diminazene	Recovered
7	Jersey	4	9.4	25.7	5.51	<i>Bbo</i> + <i>Bbi</i>	Diminazene	Recovered
8	Jersey	5	ND	ND	ND	<i>Bbo</i> + <i>Bbi</i> + <i>Bna</i>	Diminazene	Recovered
9	Jersey	4	6.8	21.9	3.88	<i>Bbo</i>	Diminazene	Recovered
10	Jersey	5	6.7	21.5	3.5	<i>Bbi</i>	Diminazene	Recovered
11	Jersey	6	2.83	10.6	2.22	<i>Bbi</i>	Diminazene	Died
12	Jersey	3	4.8	13.3	3.03	<i>Bbo</i> + <i>Bbi</i>	Diminazene	Died
13	Jersey	6	ND	ND	ND	<i>Bbo</i> + <i>Bbi</i>	Diminazene	Recovered

ND, Not done; *Bbo*, *B. bovis*; *Bbi*, *B. bigemina*, *Bna*, *B. naoakii*

(HGB: g/dl, HCT: %, RBC: $\times 10^6/\mu\text{l}$)

Cattle with severe anemia died.

Treatment and prevention

Treatment

Therapeutic dose and route of administration

Drug	Dose	Route
Diminazene aceturate	3.5 mg/kg	Intramuscular
Imidocarb	1.2 mg/kg	Subcutaneous

Prevention

Tick control

Vaccination with live-attenuated *B. bovis* and *B. bigemina* strains

Summary

1. Causative *Babesia*

B. bovis, *B. bigemina*, *B. naoakii*, and *B. divergens*

2. Lifecycle and transmission

Transmitted to cattle by specific tick vectors

3. Pathogenesis and clinical signs

Hemolytic anemia, cerebral babesiosis, and immunodisease



4. Risk factors

Age, acquired immunity, grazing management, endemic status, and breed

5. Diagnosis

Clinical, parasitological, and molecular diagnosis

6. Treatment

Chemotherapy and supportive therapy

7. Prevention

Tick control, vaccination, and chemoprophylaxis

Contact us



Dr. Thillaiampalam Sivakumar
sivavets@gmail.com



WOAH Reference Laboratory
for Bovine Babesiosis

Reference Centre



World Organisation
for Animal Health
Founded as OIE

Dr. Naoaki Yokoyama
yokoyama@obihiro.ac.jp

Visit our website for further details

<https://www.obihiro.ac.jp/facility/protozoa/en/woah-reference-centres>

Distribution, surveillance and diagnosis of JEV in animals

Dong-Kun Yang/ WOAHA expert for JE

2024. 9. 19



World Organisation
for Animal Health
Founded as OIE

Distribution of JE in human in Asia and Pacific region



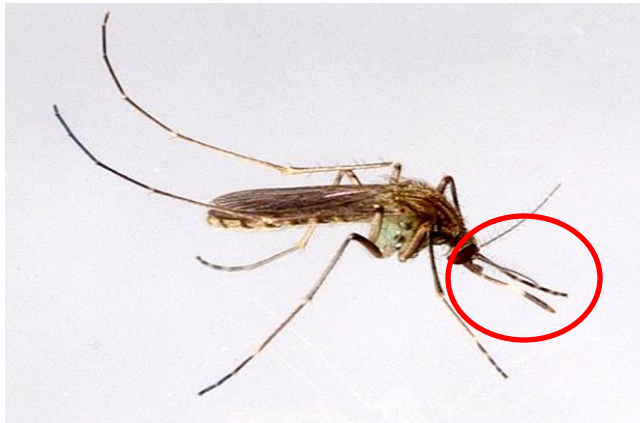
Arbovirus	Number of countries	Population (% of global population)	Estimated death per year	Chronic case per year
JEV	24	3,706,910,037 (57)	3,500 to 15,000	7,350 to 22,500

- Map of countries in Asia and Pacific region where JEV has been identified
- JEV has the **biggest population** in endemic countries among vector borne diseases

Why is JE important in animals ?

- Since the **first case of JEV infection** was reported in **1871**, **JEV infections have been reported in human and animals.**
- **Public health impact:** JE is a significant public health concern, especially in Asia and the Western Pacific regions, where it causes thousands of cases and deaths annually.
- **Neurological damage:** JE can lead to severe neurological damage, including encephalitis, seizures, and long-term cognitive and motor impairments. Approximately 20-30% of those who develop encephalitis die, and 30-50% of survivors suffer from permanent neurological sequelae.
- **Lack of specific treatment:** There is no specific antiviral treatment for JE.
- **Economic burden:** The disease imposes a significant economic burden on affected families and healthcare systems due to the high costs of medical care and long-term rehabilitation for survivors.
- **Preventable disease:** JE is preventable through effective vaccination programs and mosquito control measures.

Characteristics of *Cx. tritaeniorhynchus*, *Cx. orientalis* transmitting JEV 3 or 5



Culex tritaeniorhynchus



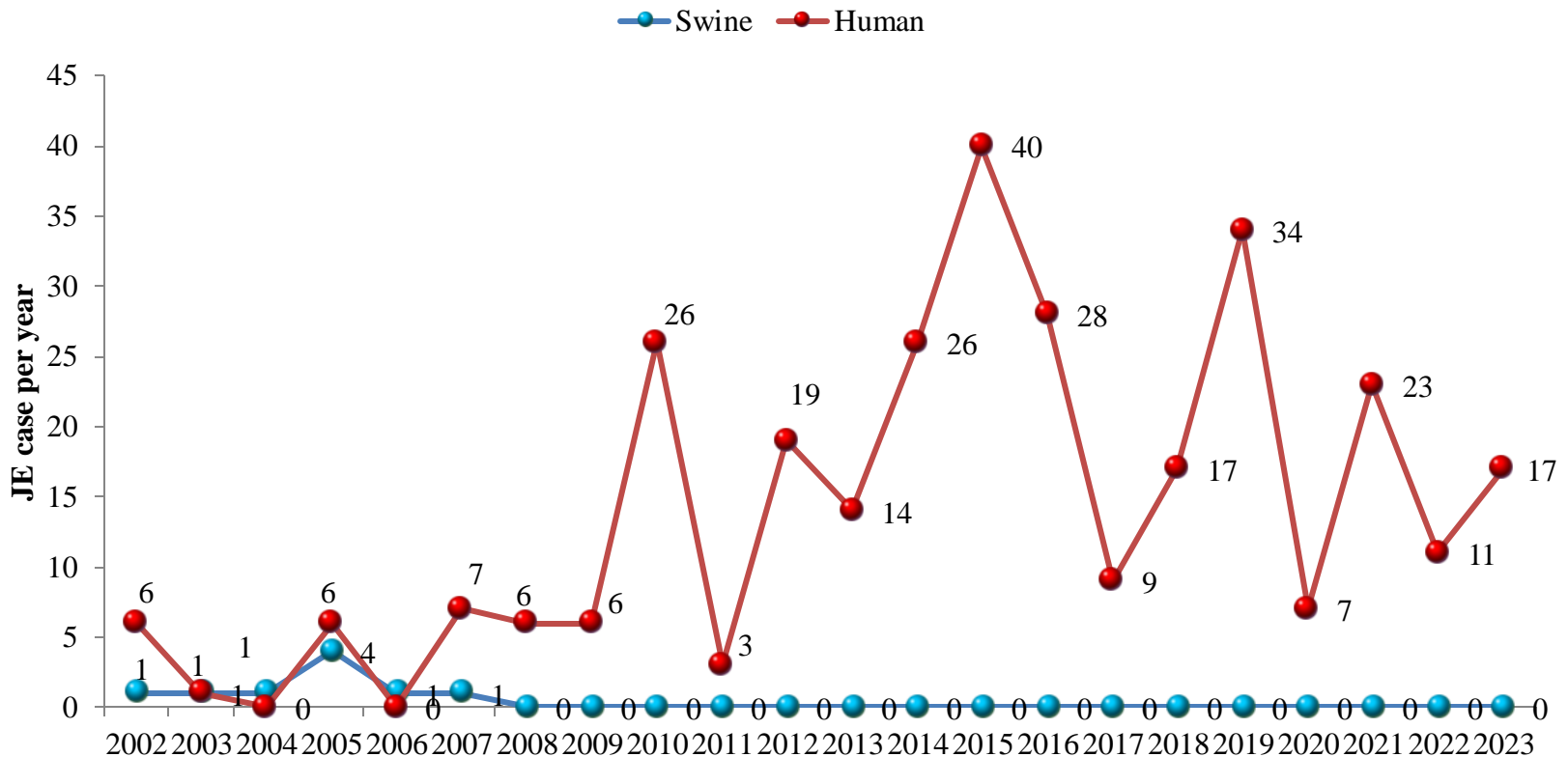
Culex orientalis

The *Cx. tritaeniorhynchus* and *Cx. orientalis* are associated with transmission of JEV genotype 3 and 5 respectively.

Habitat preference: *Culex orientalis* is commonly found in rice paddies, marshes, and other stagnant water bodies. These environments provide ideal breeding grounds for the mosquito, facilitating its role as a vector for JE.

Feeding behavior: This mosquito species is primarily zoophilic, meaning it prefers to **feed on animals rather than humans**. However, it can still bite humans, especially when animal hosts are scarce.

JE cases in human and swine in South Korea since 2002



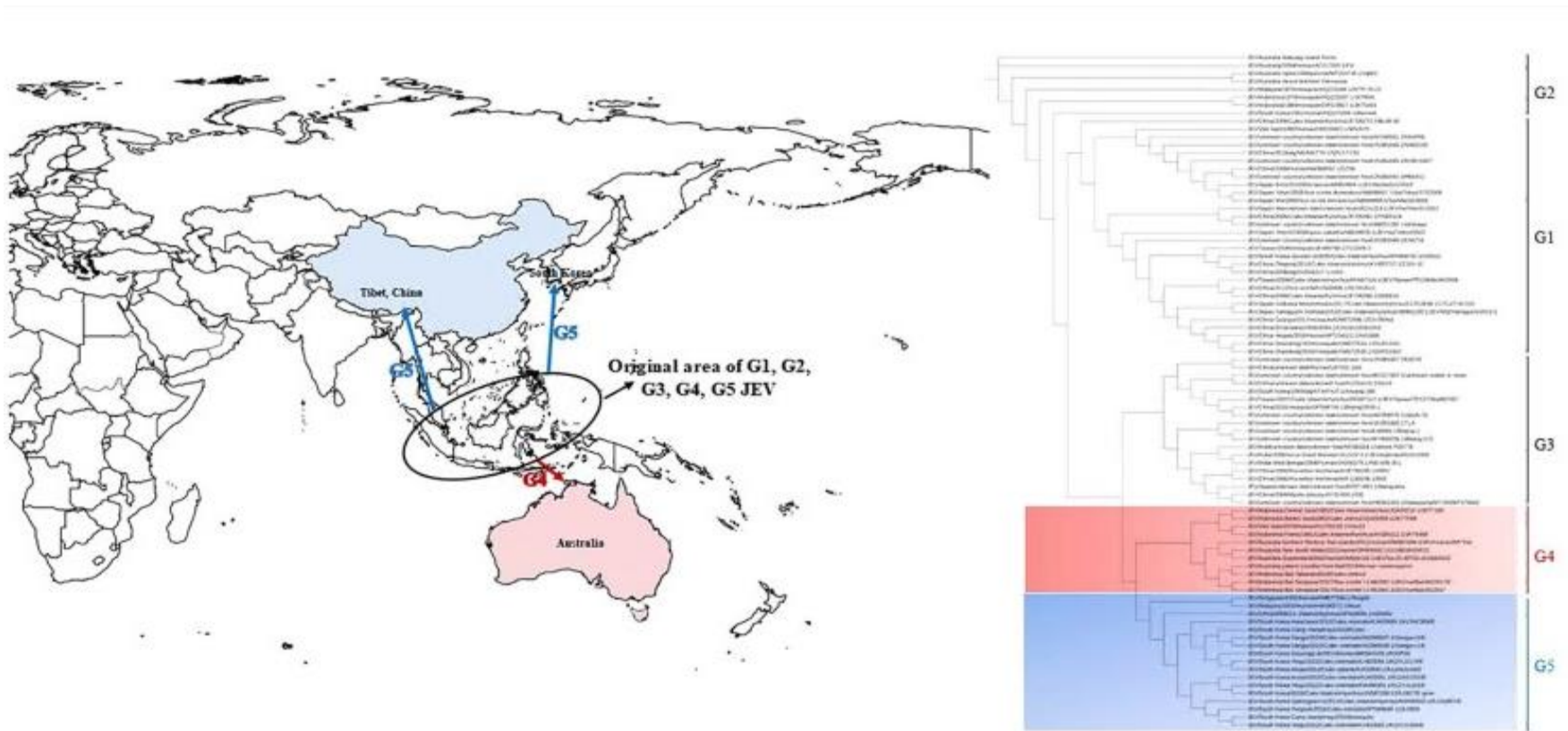
There has been no JEV infection in swine since 2008.

But, 17 JE cases occurred in human in 2023.

JEV genotype 5 has been identified in human since 2010.

JE vaccine belonging to JEV 3 has been used for Korean and animals.

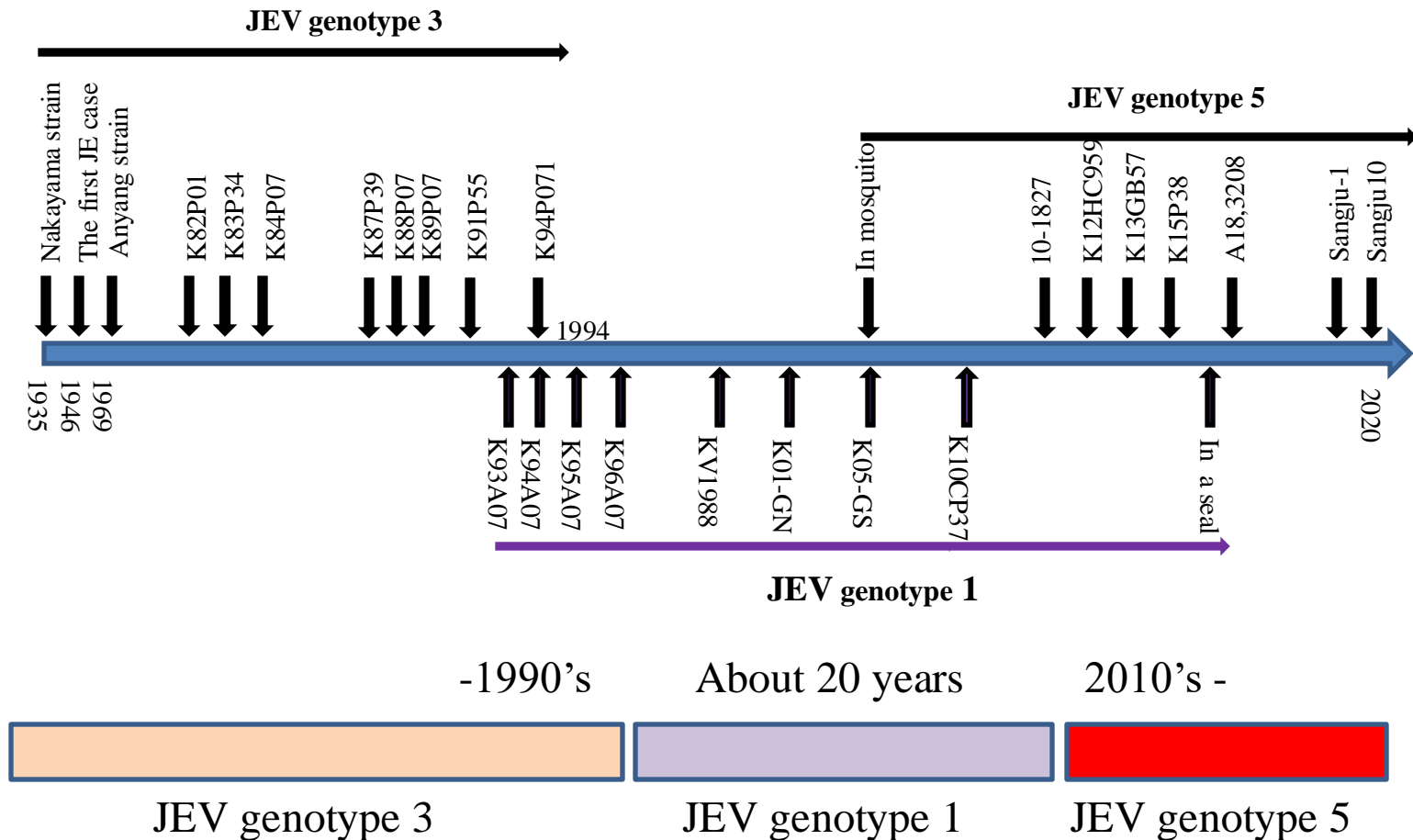
Distribution of JEV genotype in Asia in 2022



From 2009, the hidden **genotype 5** suddenly emerged from the **Tibetan** region of China and from South Korea in East Asia.

Similarly, **in 2022** the long silent **genotype 4 of JEV** emerged in Australia.

JEV genotype shift is occurring in Korea



The [detecting and monitoring the JEV carried by mosquitoes](#) in the natural environment are effective method that can help to predict whether JE genotype strains have the potential to cause human infection or not.

JEV infection was identified in a seal residing in a zoo



A seal resided in a wild zoo located in Southern region of Korea was commissioned to APQA in 2017. The seal died of heartworm and JEV infection.

The JEV was classified into **JEV genotype 1** based on the nucleotide sequence analysis.

Why do we conduct **serological survey** for JEV?

- **Early detection of antigen:** Serological tests help in the early detection of JEV in animal populations, which is crucial for monitoring and controlling potential outbreaks. This early detection allows for timely interventions **to prevent the spread of the virus to humans and other animals.**
- **Understanding transmission dynamics:** This information is vital for identifying key reservoir hosts and vectors, such as pigs and mosquitoes, which play significant roles in the virus's life cycle.
- **Evaluating vaccination programs:** Serological testing is essential for assessing the effectiveness of vaccination programs in animals. By measuring antibody levels, researchers can **determine the immunstatus of animal populations** and make informed decisions about the need for booster vaccinations or other preventive measures.

Considerations before serological test

[Pig & Horse]

1. Vaccination (Live-attenuated vaccine)
 - Vaccination policy to **sow** population in Korea
 - Vaccination to **racehorse** population
2. Maternal antibodies
3. Another flavivirus infection? [Other animals]



[Solution]

1. Vaccination or maternal antibodies : **Sampling paired sera**
2. Other *flavivirus* infection : **Cross neutralization test**
 - e.g. JEV \longleftrightarrow WNV(exotic) : difference above 4 fold dilution

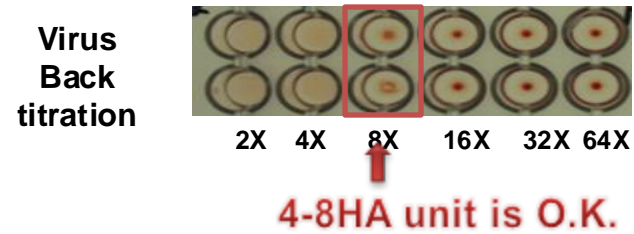
How to conduct HI test



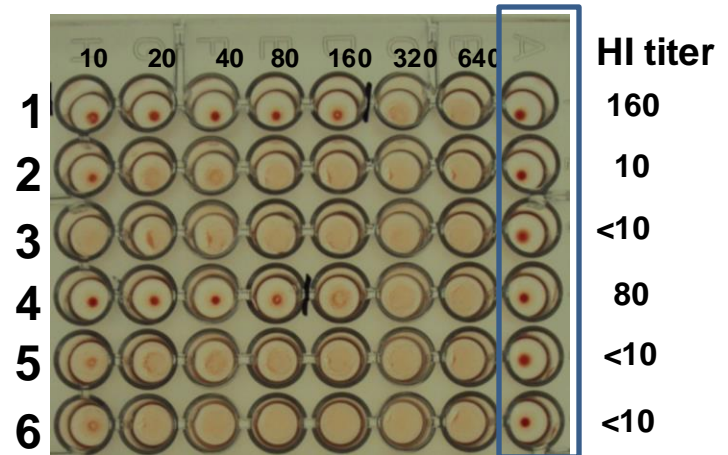
The commercial HI kit for JEV



Goose red blood cells packed 100%

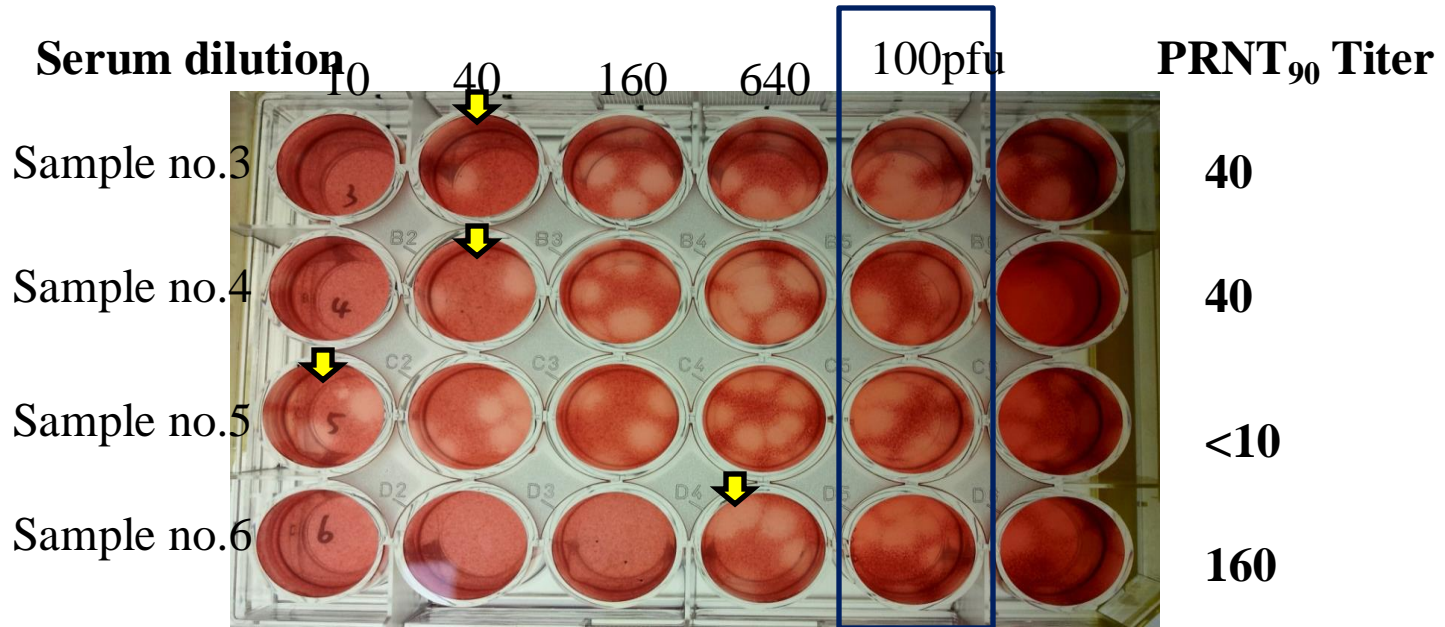


Sample no.



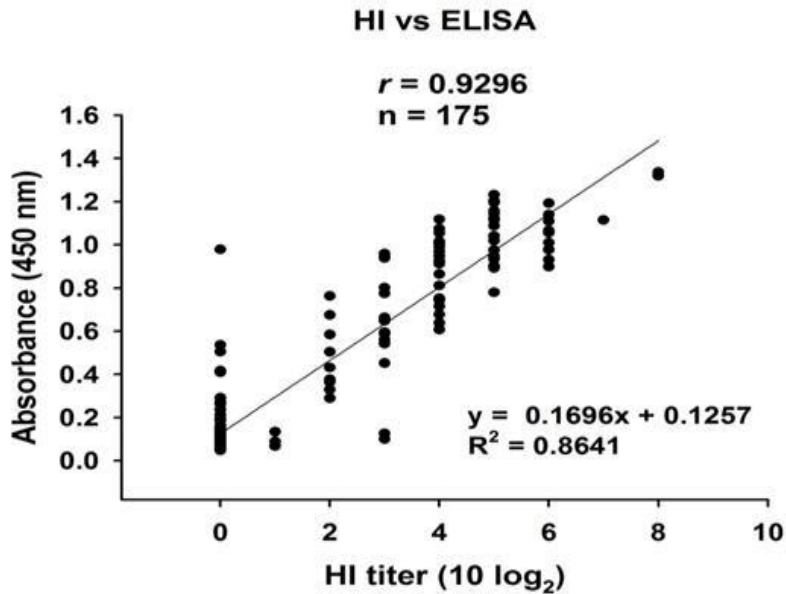
The [commercial HI kit](#) for the detection of JEV antibodies is available for animals.

Example of PRNT to detect JEV antibodies



- The principle of plaque assay is to differentiate accumulated dead cells by virus infection from surrounding surviving cells.
 - PRNT₉₀ means 90% of JEVs were inhibited by antibody within serum.
- There are some **limitations of PRNT** in testing large numbers of sera.

Correlation between ELISA and HI test

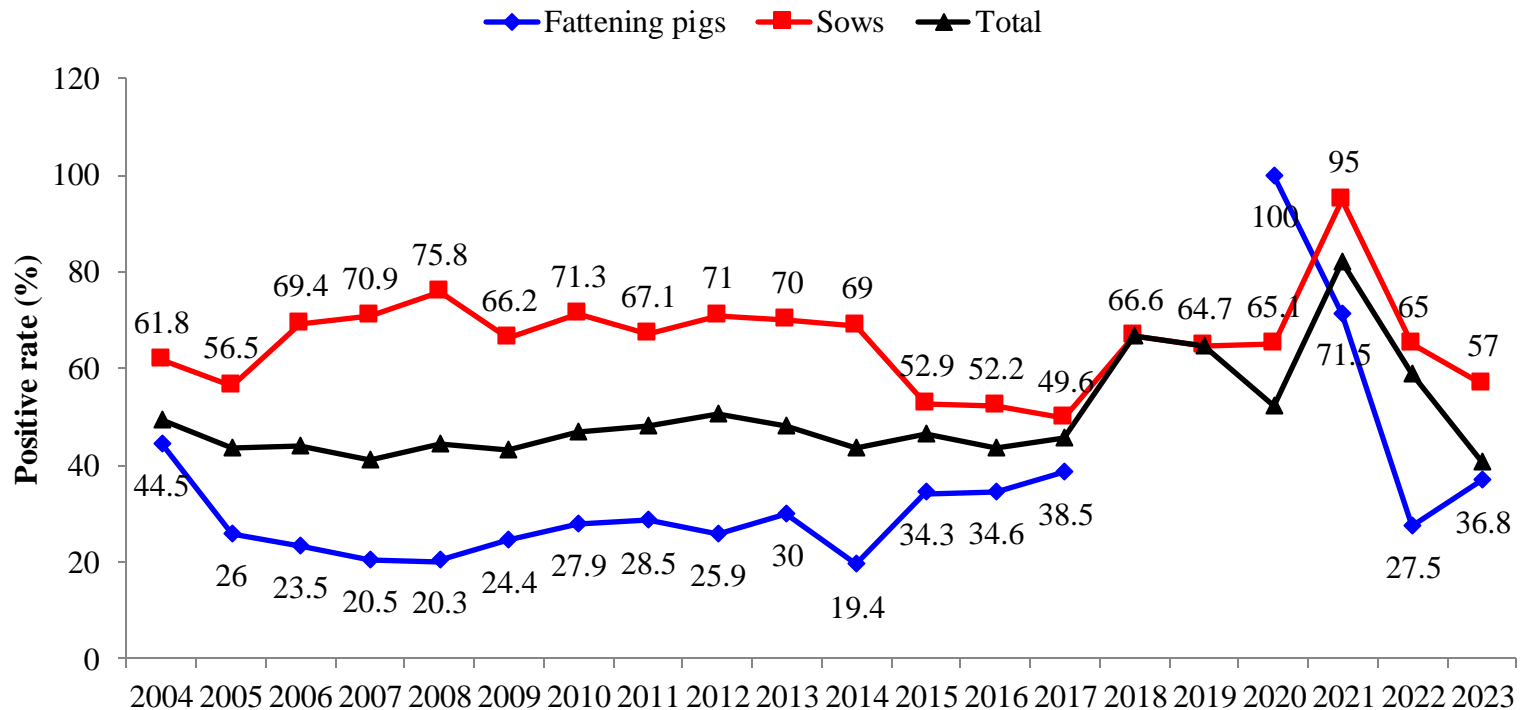


N=175 sera		No. of samples with HI	
		Positive	Negative
ELISA	positive	71	8
	Negative	6	90
	Sum	77	98
Sensitivity		92.2%	
Specificity			91.8%
Accuracy		92.0%	



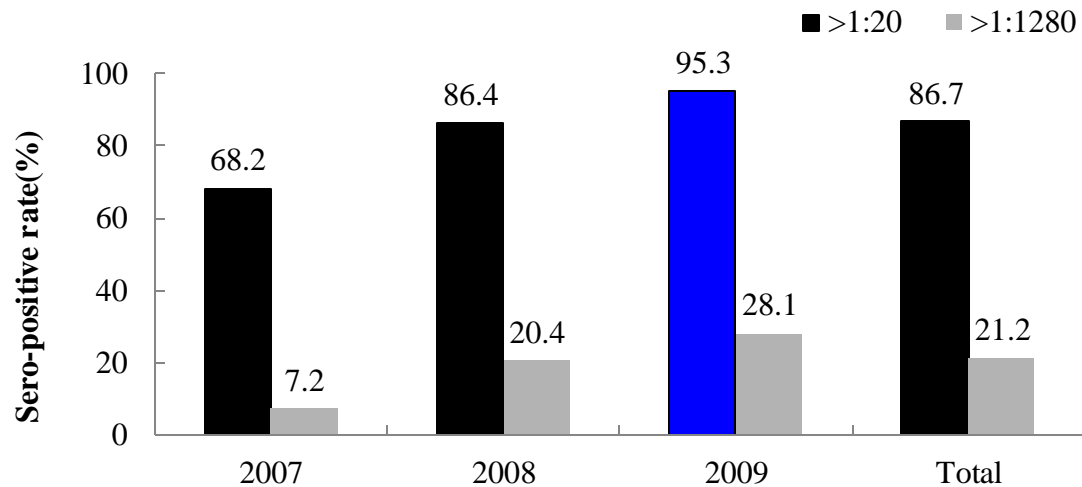
- These results suggest that **I-ELISA** is useful for sero-surveillance of JEV in swine.

Sero-surveillance of JEV in pigs since 2004



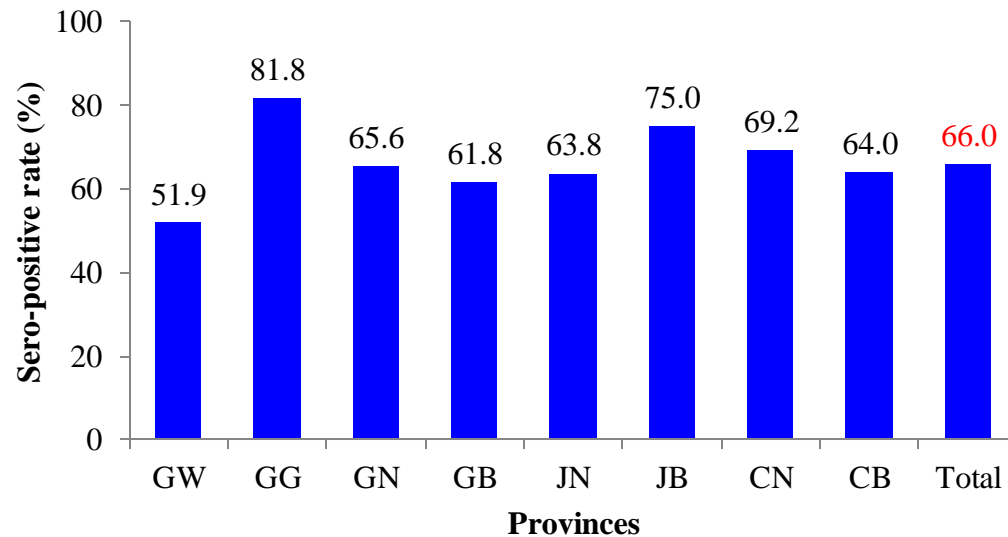
- Sera obtained from 5,000 to 8,000 pigs each year have been checked by HI or ELISA test since 2004.
- HI and ELISA tests have been used to measure JEV antibodies in pig sera since 2017.
- **If all sows on a pig farm are negative for JEV, vaccination is recommended.**

Sero-surveillance of JEV in **wild birds** captured in South Korea



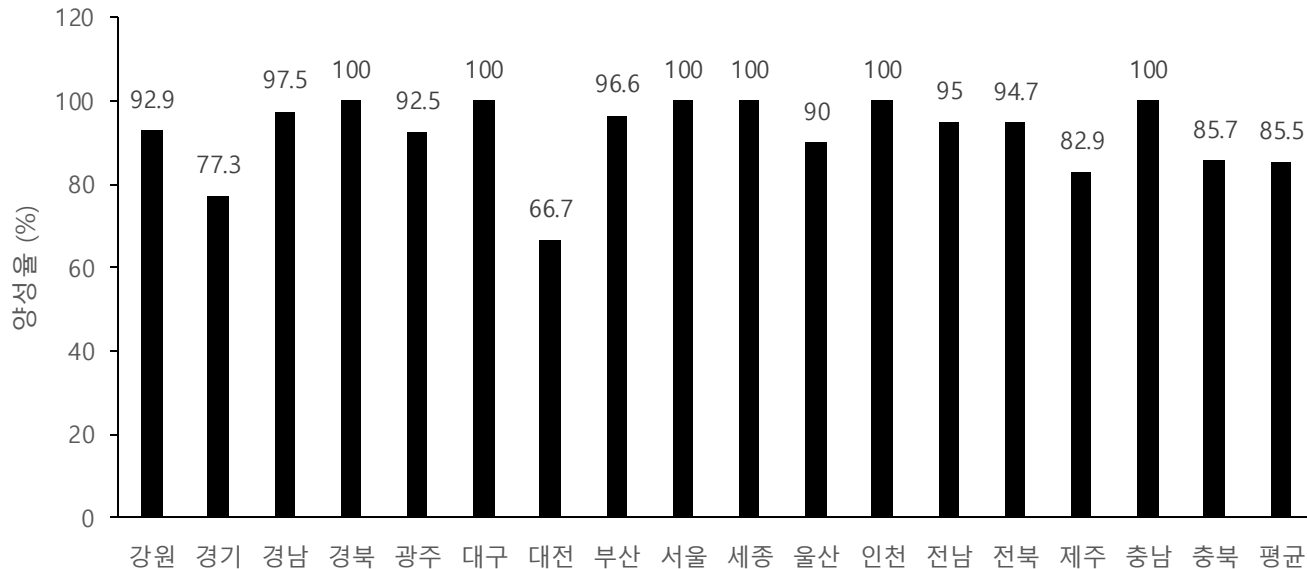
- Blood samples were collected from **1,316 wild birds** including migratory birds in 16 sites of 6 provinces.
- Of the 1,316 serum samples tested, 1,141 sera (**86.7%**) were positive for JEV.
- Migrating birds are believed to be one of the factors responsible for transmitting the JEV.
- When the chicken is sero-positive against JEV, it means the widespread presence of JEV.

Sero-surveillance of JEV in **wild boars** in South Korea



- The results showed that **66.0% (190/288) of wild boars** in Korea had neutralizing antibodies against JEV.
- Wild boars are amplifying hosts of the JEV and are believed to be one of transmitters.

Sero-surveillance of JEV in horse sera



- Of the 1,331 horse sera collected in 2023, **85.5%** were positive for JEV.
- As a result of analyzing regional distribution, the number of horses in **Daejeon** was the lowest at 66.7%.
- The JEV antibodies in horse are an immune response following vaccination and JEV vaccine is recommended **for antibody-negative horses**.

Overview of **diagnosis** of JEV antigen in animals

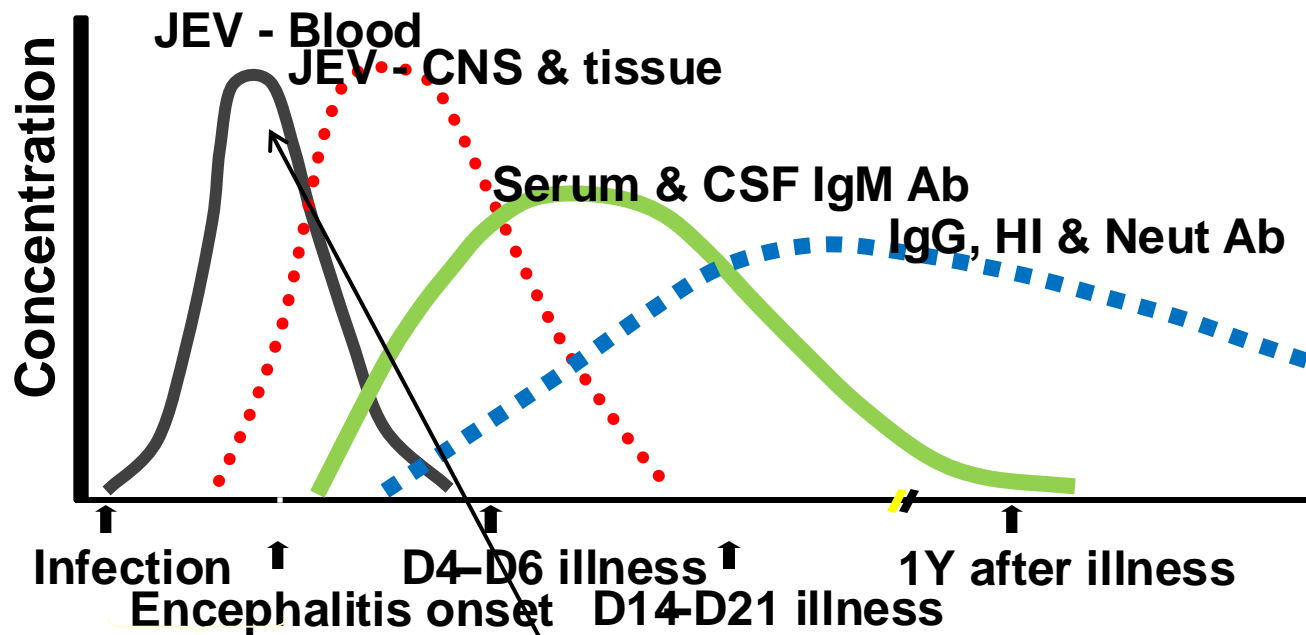
Method	Purpose					
	Population freedom from infection	Individual animal freedom from infection prior to movement	Contribute to eradication policies	Confirmation of clinical cases	Prevalence of infection – surveillance	Immune status in individual animals or populations post-vaccination
Detection of the agent¹						
Virus isolation	–	–	–	+++	–	–
Antigen detection	+	+	+	+	+	–
Real-time RT-PCR	++	++	++	+++	++	–

- Diagnostic test methods including virus isolation, real time RT-PCR, mouse inoculation have been used to detect JEV antigens.

Diagnosis of JE in pigs and horses

- Most JE infection in sows manifests as a **reproductive disease** to reach 50–70% **abortions in sows, subclinical in horses**.
 - Stillbirths or mummified fetuses; usually at term
- Live **born piglets** most often demonstrate **neurologic signs** of tremors and convulsions and may die soon after birth.
 - Mild febrile disease or subclinical disease in non-pregnant females
 - Natural infection results in long lasting immunity
 - **Mortality rate is near zero in adult swine**
 - **Identification of JEV on brain or spinal cord in horse**
 - **Under 70-day old fetus: JEV antigen has to be identified with VI, or RT-PCR.**
 - **Over 70-day old fetus: JEV antibody on thoracic fluid should be checked.**

Host response to JE infection



Virus strain	Route	ID. of pig	Post inoculation days						
			2	4	6	8	15	22	
KV1899-37P	IM*	1	-/-	-/-	-/-	-/-	-/-	-/-	
		2	-/-	-/-	-/-	-/-	-/-		
		3	+/+	-/-	-/-	-/-	-/-		
		4	+/+	-/-	-/-	-/-	-/-		
Control	-	1	-/-	-/-	-/-	-/-	-/-		

+ / +: Positive results in both **virus isolation and real-time RT-PCR**.

Because of short period of viremia, the timing of virus detection must be considered.

Diagnostic **procedure** to detect JE antigen in suspected samples

- Cerebro-spinal fluid, **brain tissue**, aborted fetus, plasma, **Mosquito** etc

Antigen preparation(10% homogenization etc) and **extraction of RNA**



One step RT-PCR or real time RT-PCR
(Target : E or NS gene)

Negative

The End

Positive

Virus Isolation **from sample material**
in C6/36, BHK-21, and Vero cells



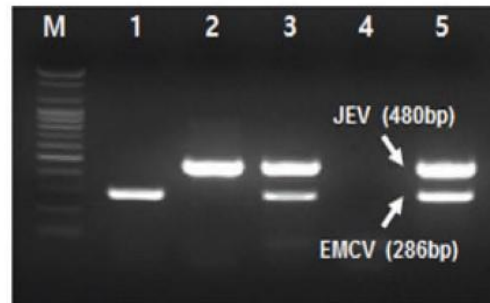
sequencing

Characterization

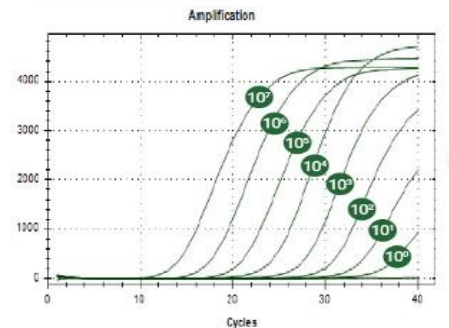
(IFA, EM, Inoculation in suckling mouse, HA etc)

Multiplex RT-PCR and real time RT-PCR kit to detect JEV

Abortion MP RT-PCR II



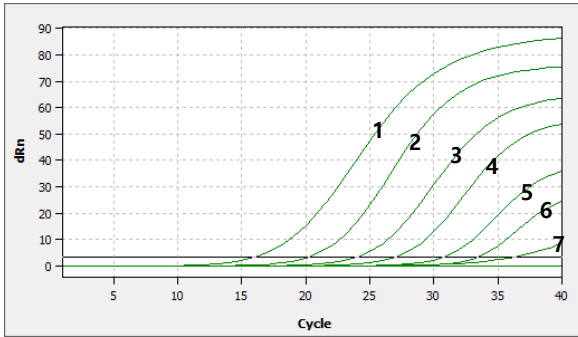
JEV (HEX)



- **Conventional RT-PCR kits** for the detection of JEV in brain, fetal fluid and sera have been commercialized.
- **Real time RT-PCR kit** is also available to detect JEV in samples.

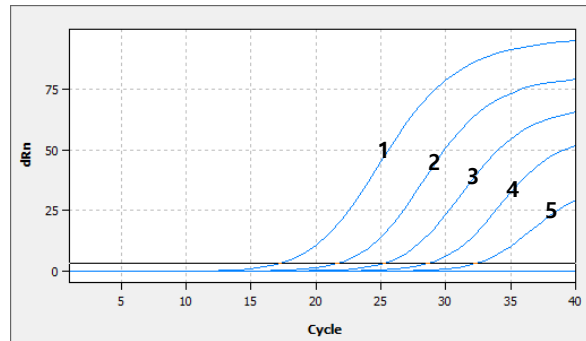
Example of application of real time RT-PCR kit to JEV genotypes

JEV G1



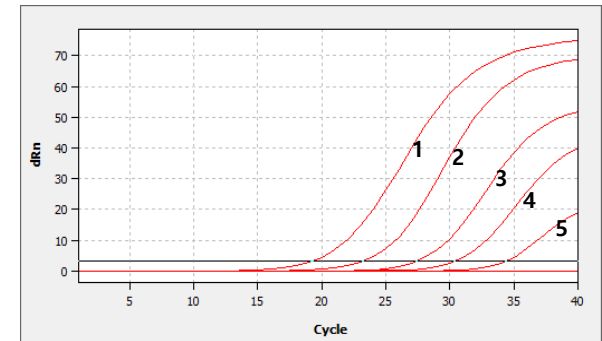
1.18 TCID₅₀/mL

JEV G3

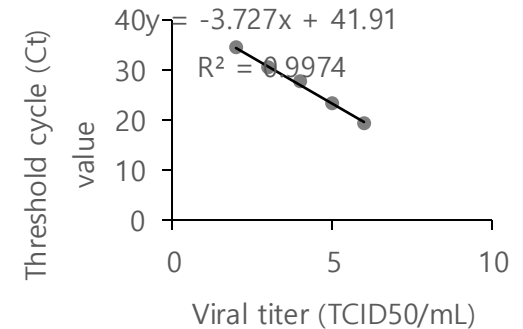
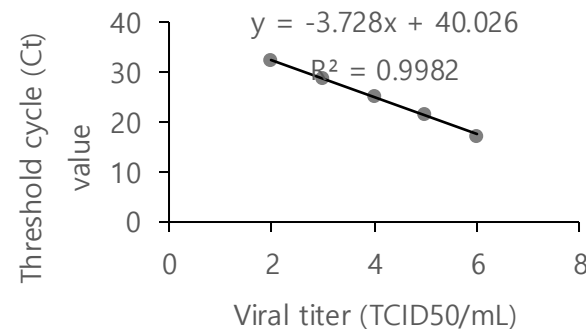
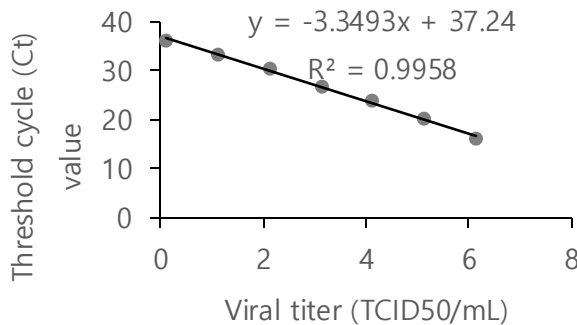


6.48 TCID₅₀/mL

JEV G5

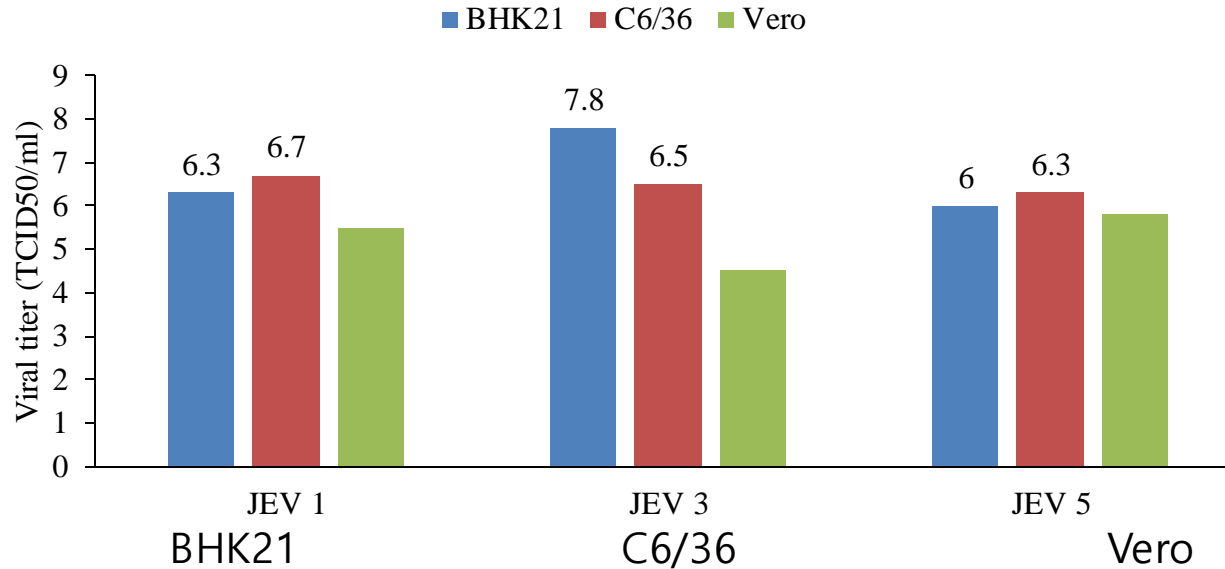


20.77 TCID₅₀/mL

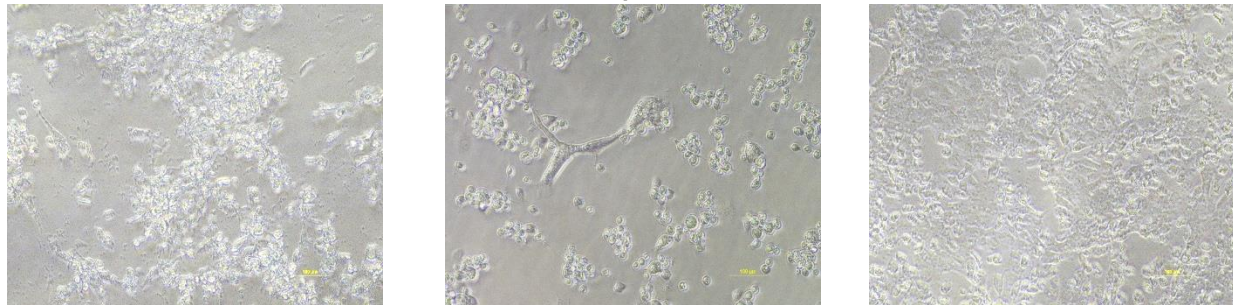


- The real time RT-PCR kit detected three genotypes of JEV and showed the lowest detection limit to JEV genotype 1 of 1.18 TCID₅₀/ml.

Proliferation ability of JEVs in 3 types of cells



JEV 5



- JEV has different proliferative abilities depending on the cells.
- **BHK-21 and C6/36 cells** are suitable for isolating JEV.
- C6/36 cells should be culture at 28 degrees.

Mouse inoculation test to detect JEV



- **The 3-5 day old nursing mice** inoculated with JEV showed paralysis, signs of nerve system and died within 7 days post inoculation.
- **Four-week old mice** inoculated with JEV via intracranial route died of neurologic disease. The most susceptible five mouse species to the JEV are [C3J/He](#), [DBA/2](#), [C57BL/6](#), [Balb/c](#), [ICR](#).

How to control JE in animals

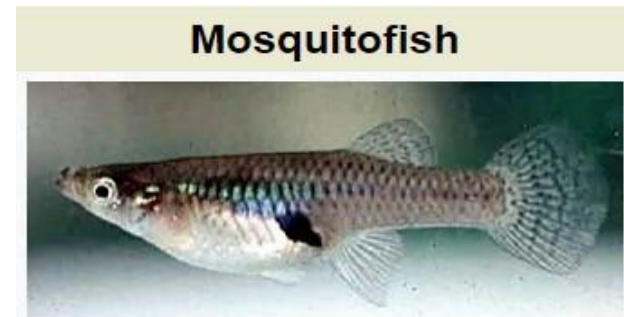
- **Vaccination:** Implementing vaccination programs for animals, especially pigs, **can significantly reduce the spread of JEV**. Vaccines help build immunity in animal populations, preventing the virus from amplifying and spreading.
- **Mosquito control:** Reducing mosquito populations through environmental management, such as **eliminating standing water** where mosquitoes breed, and **using insecticides** can help control the primary vectors of JEV.
- **Active surveillance:** Regular surveillance and monitoring of animal populations for signs of JEV can help in **early detection** and prompt response to outbreaks. This includes serological testing to identify infected animals.
- **Public awareness and education:** Educating farmers and animal handlers about the importance of mosquito control, vaccination, and early detection measures can enhance the effectiveness of control programs. Awareness campaigns can also promote the adoption of best practices to prevent the spread of JEV.

JEV vaccination and vector control

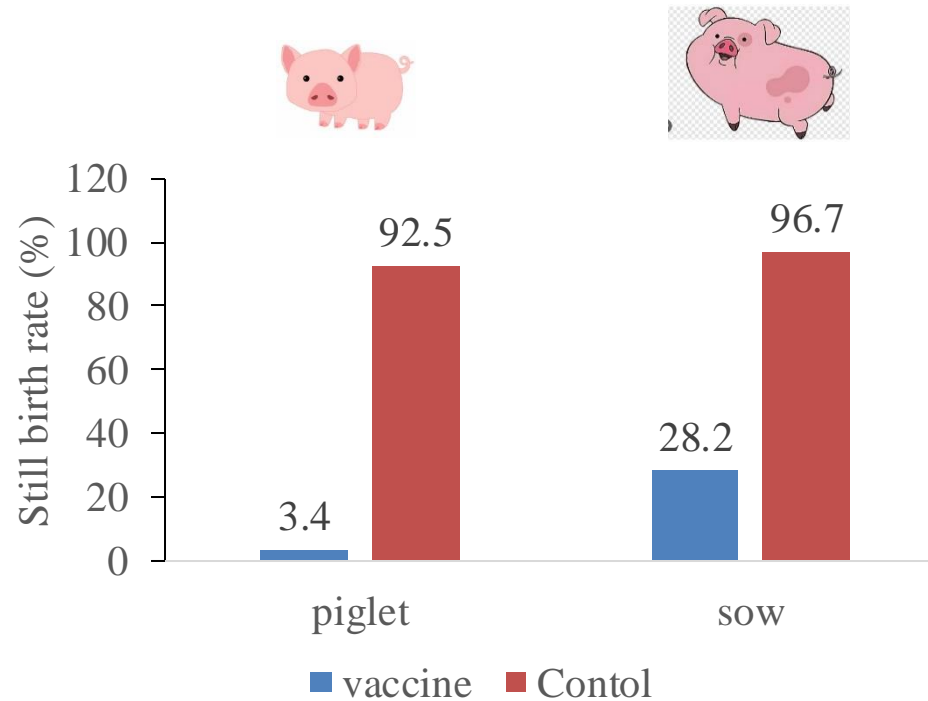
- **Vaccination** is the most effective preventive method for animals.
- **Minimizing exposure** of animals to mosquitoes using nets is possible.

Type	Virus strain	Culture method	For use in	Genotype
Inactivated JE	Nakayama	Chicken embryos	Horse/pigs	3
Inactivated JE	Beijing	-	Horse/pigs	3
Live JE	AT	Hamster kidney cell	Pigs	3
Live JE	M	Hmlu-1	Pigs	3
Live JE	Anyang300	Duck primary cell	Pigs	3
Live JE	SA-14-14-2	BHK-21 cells	human and Pigs	3
Live JE	M-17	Porcine kidney cell	Pigs	3

- **Vectors** can be controlled by using **insecticides** in rice fields, growing **larvivorous fish and mosquitofish**.



Example of JEV vaccine application on pig farms



- In total, 3,790 sows were inoculated with JEV vaccine in three provinces in 1975.
- The stillbirth rate in sows inoculated with JEV vaccine was reduced rapidly.
- But sows that were not treated with JEV vaccine showed high still birth rates (92.5 to 96.7%).

Summary

- **Continuous cooperation and efforts** are needed to improve human and animal health.
- **Strengthen the monitoring of JEV gene changes:** it is essential to enhance methods for detecting and monitoring JEV genotypes in the natural environment. In Europe and Africa, JEV infections were reported.
- **Enhanced surveillance for early detection:** as mosquito activity is expanding due to climate change, active surveillance into JEV is required in Asian countries.
- **Vaccination:** continuous vaccination of pigs and horses is recommended to prevent JEV infection in animals.
- **Development of new JE vaccine:** the JEV 3 vaccine has the protection rate against JEV 1 ranging from 92% to 97%.

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T E R A M O

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West Nile Fever

WOAH Regional Workshop on Vector Borne diseases in Asia and the Pacific

Federica Monaco

f.monaco@izs.it

*Istituto Zooprofilattico Sperimentale
dell'Abruzzo e del Molise "G. Caporale"*

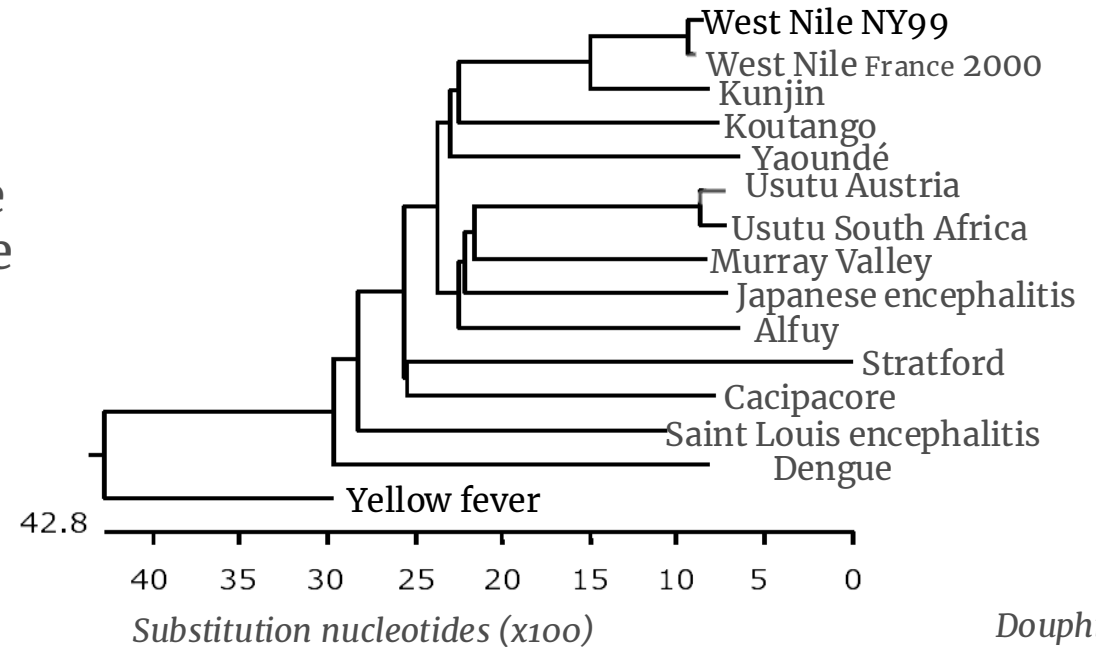
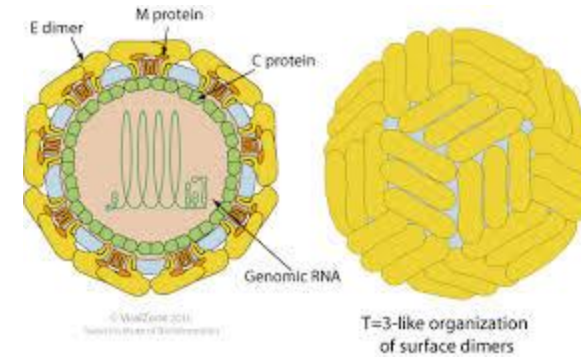
WOAH Reference laboratory for WNF

Tokyo, 19th September 2024

West Nile virus (WNV) is a mosquito-borne virus belonging to the genus *Flavivirus* in the *Flaviviridae* family capable of infecting a wide range of species

Serologically, West Nile virus is a member of the **Japanese encephalitis serocomplex**

West Nile virus

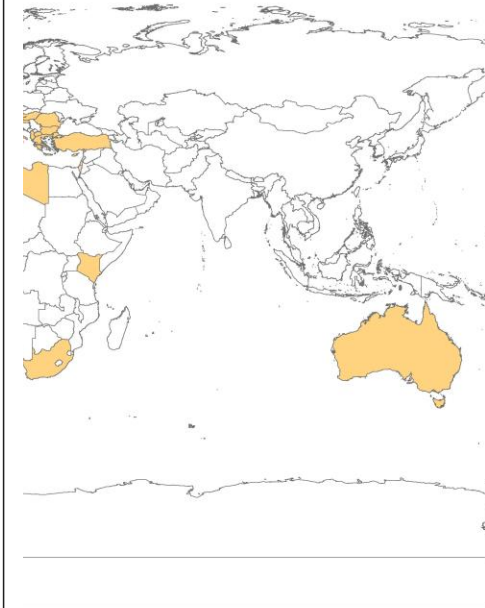
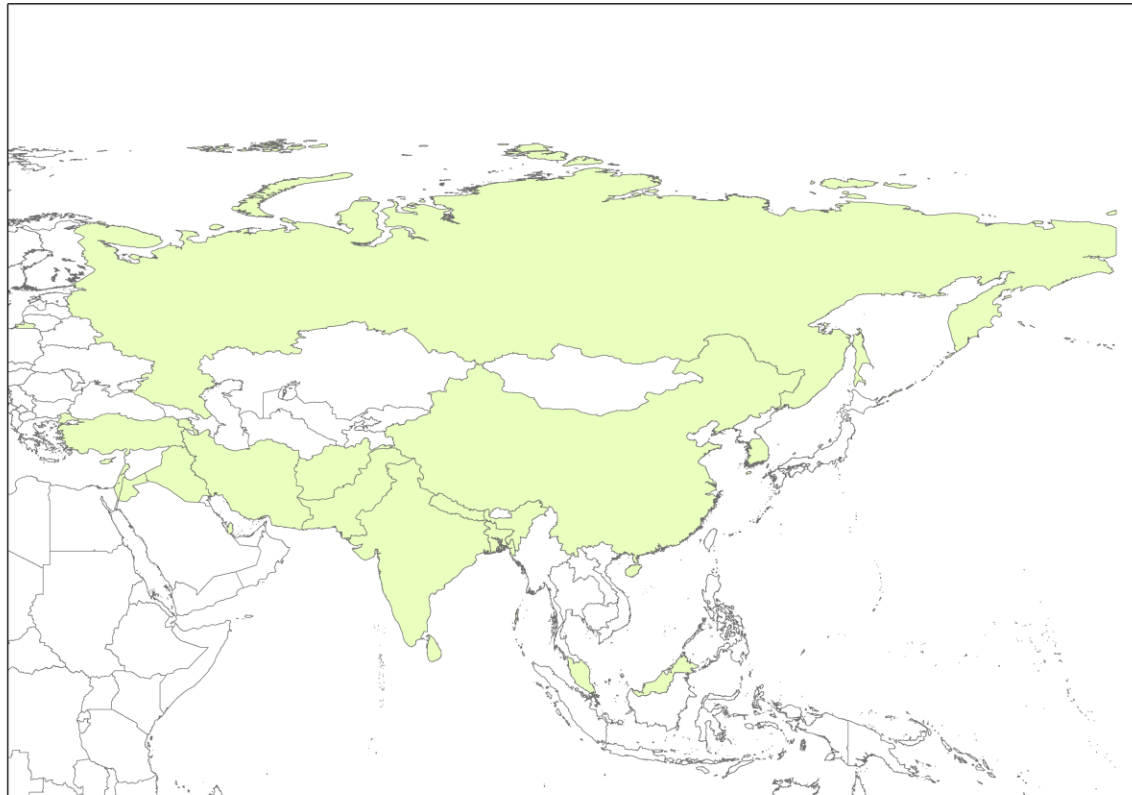


Douphin, 2006

West Nile virus

WNV is among the most widespread flaviviruses originally identified in 1937 and has been reported in Africa, Europe, North America, and Asia.

Several WNV lineages have been identified, with lineage 2 being the most responsible for the current outbreaks in Europe.



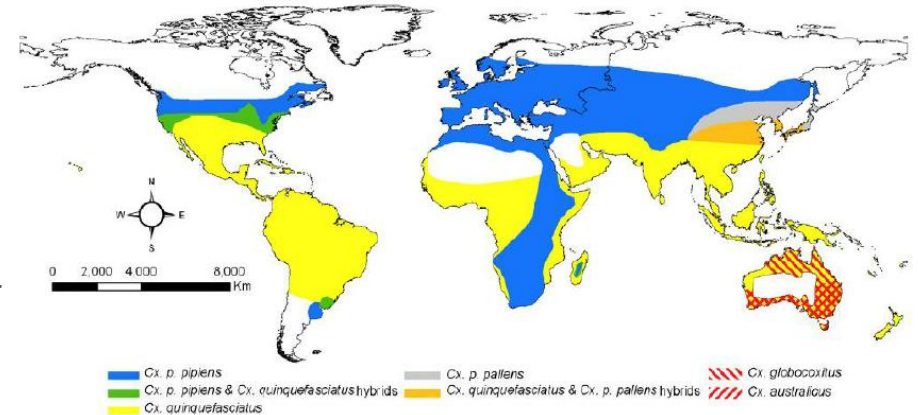
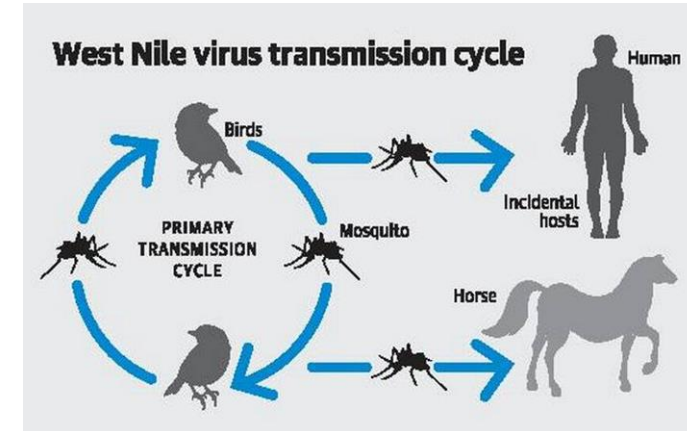
Data source: WAHID and ECDC
(2019-2024)

WNV is maintained in nature by enzootic cycle between **adult ornithophilic mosquitoes** in mainly belonging to the **Culex**, genera and several bird species

WNV is predominantly vectored worldwide by members of the **Culex pipiens complex**

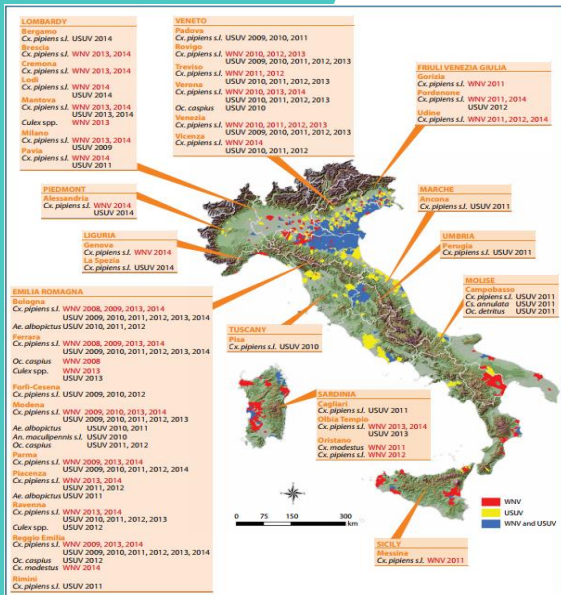
Horses and **humans** are considered dead end hosts of the virus and do not contribute to the transmission cycle

West Nile virus



Global distribution of *Cx. pipiens* complex mosquitoes. Geographic range for *Cx. p. pipiens* includes both forms (*pipiens* and *molestus*). *Cx. australicus* and *Cx. globocoxitus* are restricted to Australia.

(Ciota and Kramer 2013)



The **incubation period** is 2-14 days

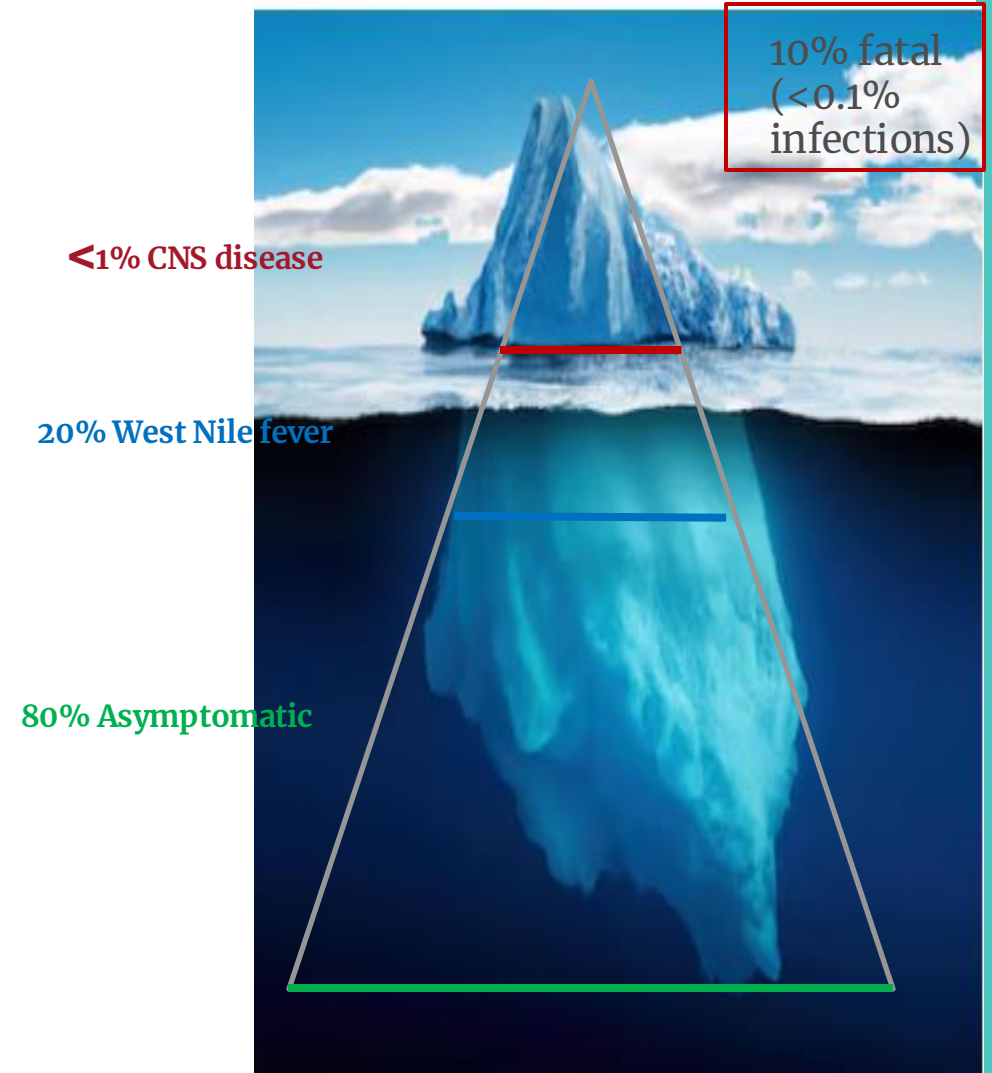
80% of human infections have no symptoms

About **20%** of WNV infections in humans may cause West Nile fever (WNF),

Less than **1%** may cause West Nile neuroinvasive disease (WNND) that affects the nervous system.

1 WNND \longrightarrow **150 infections**

Symptoms in human



The **incubation period** is 3-15 days

The infection usually is **asymptomatic** (>70%) or with **mild symptoms** like fever, weakness, myalgia (20%):

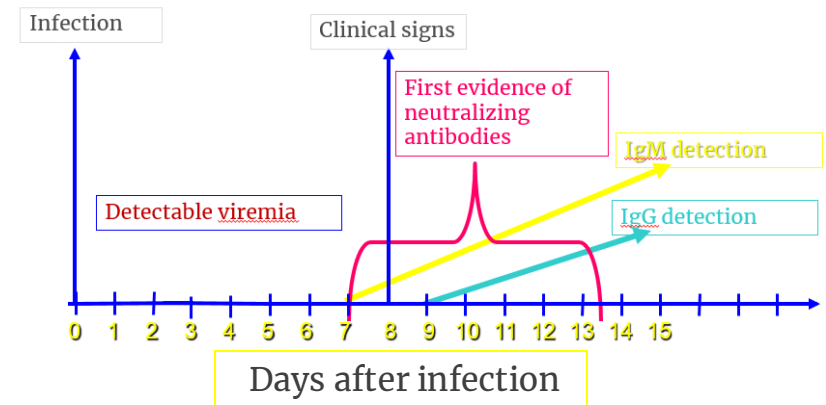
- 1-10% of infected horses has the **neuroinvasive form**
- virus in the CNS (spinal cord, hindbrain, midbrain)

The **fatality rate** in horses can reach 57-60%

- In USA (2000) was 38.3%
- In Italy was 42% (1998), 15.6% (2008)
24.3% (2009)

Viremia precedes the clinical manifestations of disease
Rarely, horses are symptomatic AND viremic

Symptoms in horses



Bunning et al., 2002

Serum, blood,
tissue, mosquitoes

Real time RT-PCR (OIE, 2018)

NEG



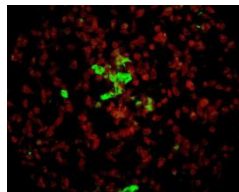
IV (C6/36, Vero,
BHK21, RK13)

POS

Ct threshold

WGS

(Diagne, 2023)



Identification (IF or RT-PCR)

WGS

(Diagne, 2023)



Serum

ELISA IgG & IgM

POS

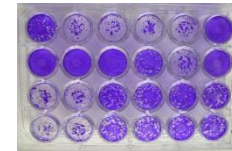
PRNT/VN

(OIE, 2018)

WND

Usutu

TBE ...



NEG

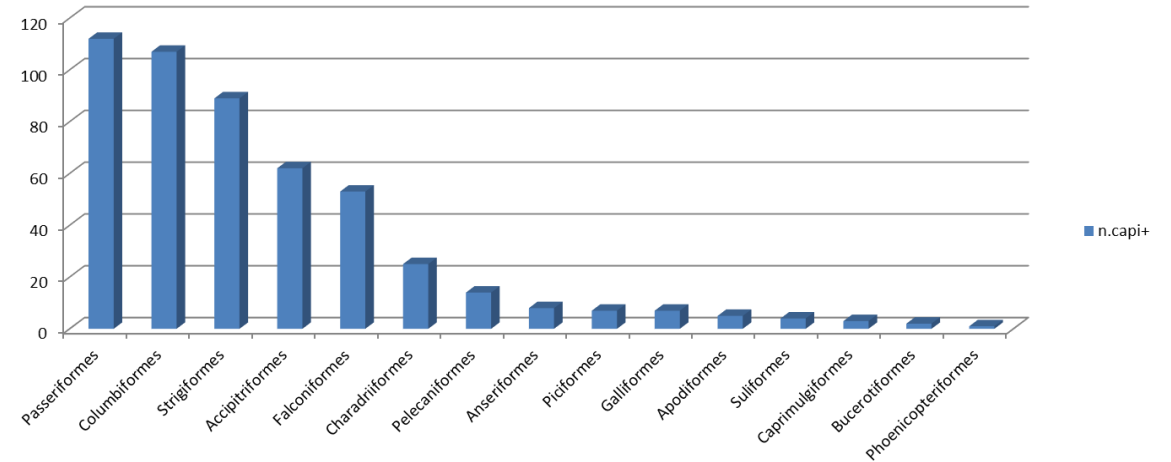
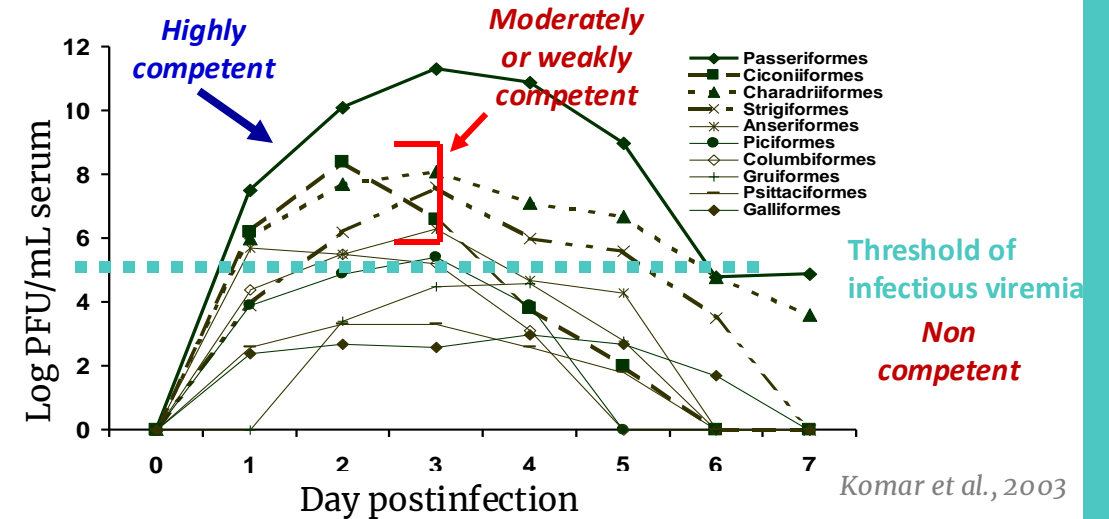


Over **300 species** of birds have been identified as viable WNV hosts

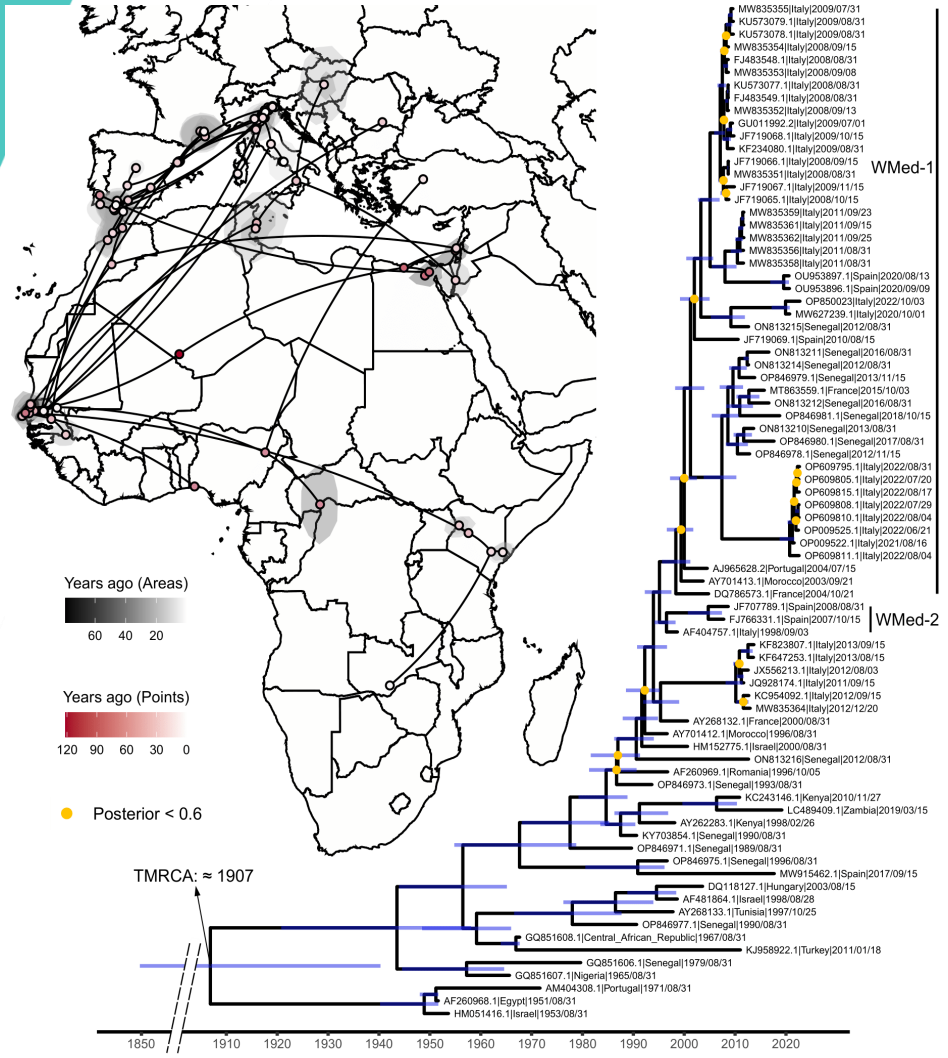
There is significant variability in **viremia levels** and **disease**

Clinical signs following WNV infection is uncommon and related to bird **species** (birds of prey) and **viral strain**

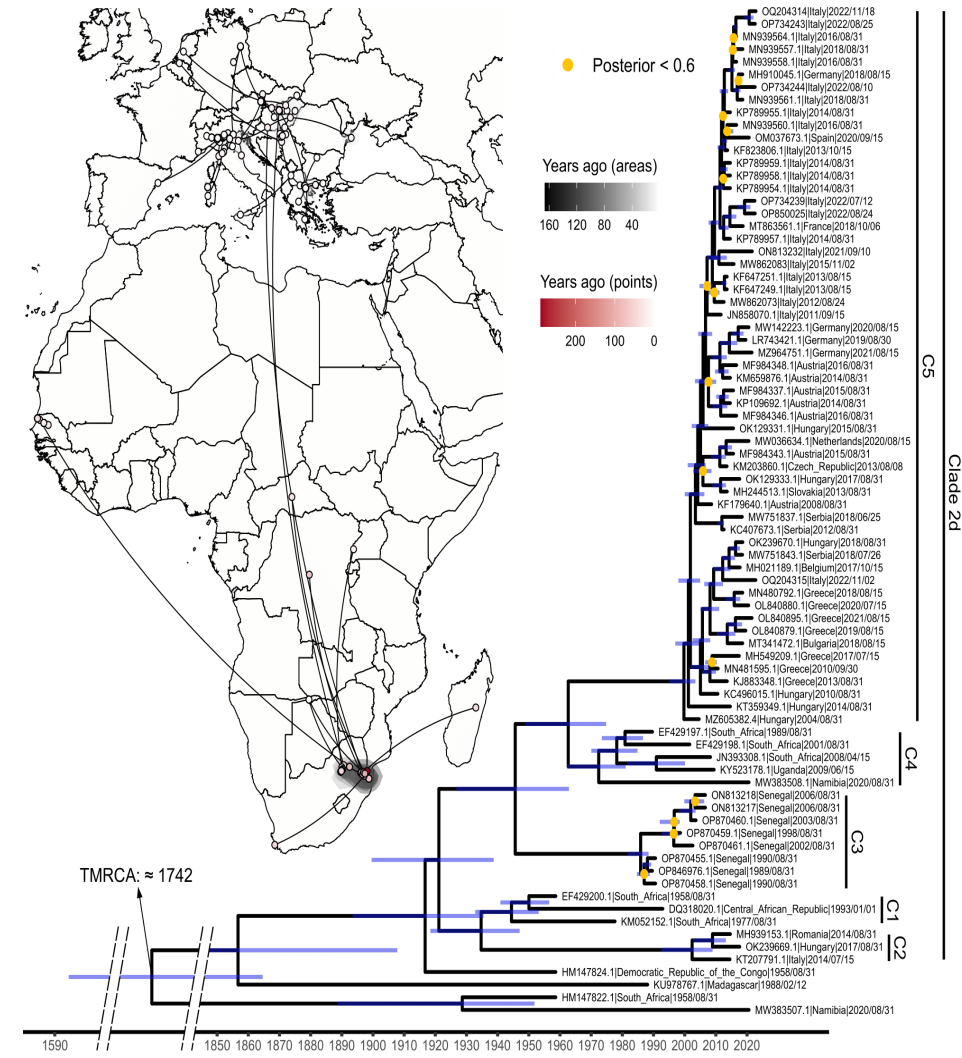
Birds



WNV L1



WNV L2



Transmission - SoHO

The virus can also spread between humans through **blood transfusion** and **organ transplant**. Other ways in which the virus can spread are **from mother to child during pregnancy** and **breast-feeding** and through **laboratory exposure**

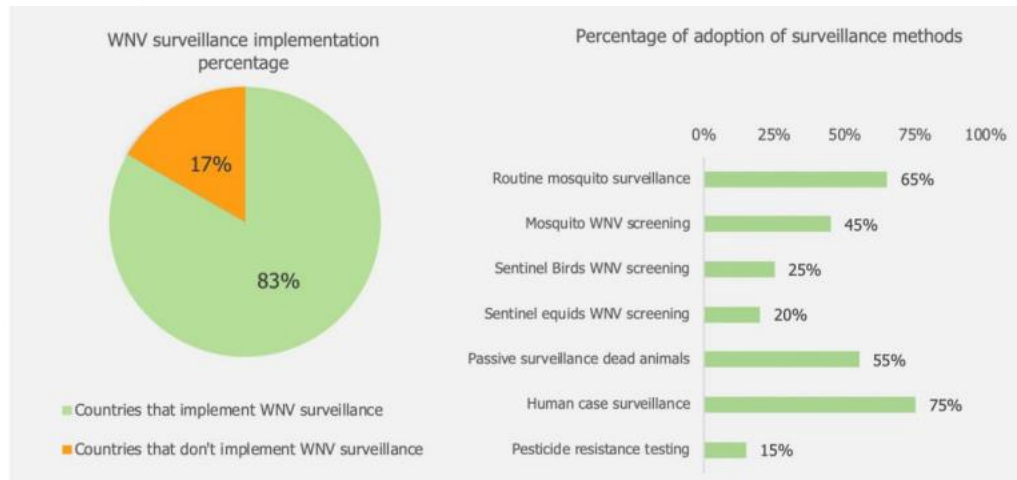


Importance of veterinary surveillance

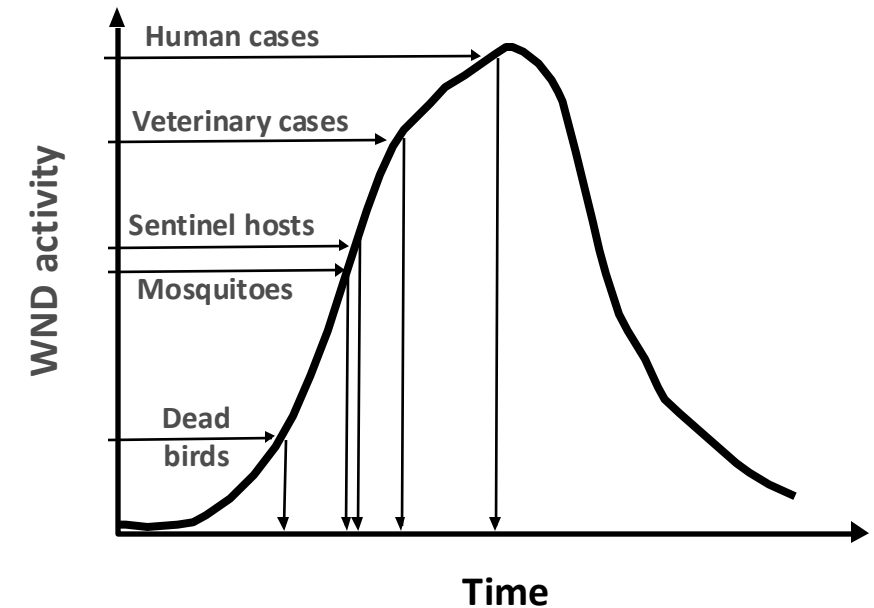
Early detection of WNV circulation for rapid risk assessment and adoption of appropriate preventive measures in **public health**.

Surveillance of WNV circulation requires a **multidisciplinary effort**

- complexity of the epidemiological cycle



ECDC, 2020



Entomological and veterinary surveillance are keys:

- to assess the associated **human health risk**
- to trigger a more timely and effective **control of the disease** in humans

WNV surveillance: the Italian model

High transmission risk area

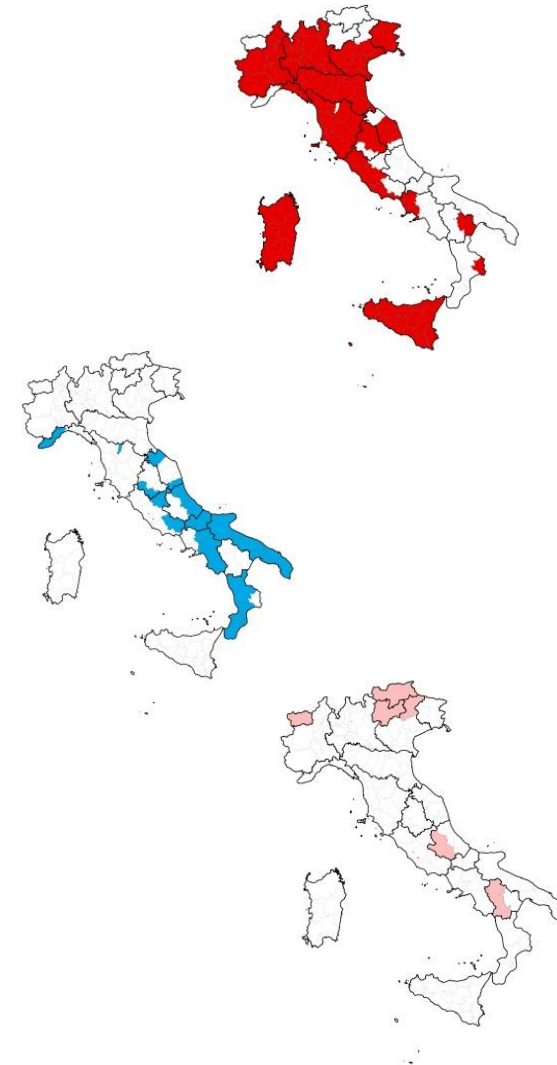
Territories (Provinces NUT-3) where WNV is circulating or has circulated in at least one of the 5 years before as well as the surrounding areas

Low transmission risk area

Territories (Provinces NUT-3) where WNV has never been/rarely reported, which have eco-climatic condition favorable to viral circulation

Minimum transmission risk area

Territories (Provinces NUT-3) where WNV has never been reported and where eco-climatic conditions are not suitable to WNV circulation



WNV surveillance: the Italian model

Veterinary surveillance is focused on the following components:

- ✓ Surveillance of resident birds of target species (**Magpie, Carrion Crow and Eurasian jay**) in **High** and **Low risk** areas
 - ✓ alternatively in Low risk areas surveillance can be conducted on rural or open air poultry rearing units.
- ✓ Entomological surveillance. **High** and **Low risk** areas (1 trap/20X20 km)
- ✓ Horses clinical surveillance. (**whole country**)
- ✓ Wild bird mortality surveillance (**whole country**)



To guarantee harmonization and representativeness of the data collected within active surveillance each province (NUT-3) is divided in units of 1200-1600 km² and activities referred to each unit.

Official veterinarian

coordinated on a regional level

implemented surveillance activities

collect each type of species (bird, horse, mosquitoes)

notify suspected in SIMAN

Send samples to IIZZSS

notify confirmed outbreaks in SIMAN

T E R

ISITUTO ZOOPROFILATTICO SPERIMENTALE DELL'ABRUZZO E DEL MOLISE "G. CAPORALE"

WN infection clinical suspicion/ surveillance activities within National Plan

IIZZSS

at regional level

tested each type of samples/species (bird, horse, mosquitoes)

In case of positivity, send the samples to CESME

CESME

At national level

Confirms suspected specimens

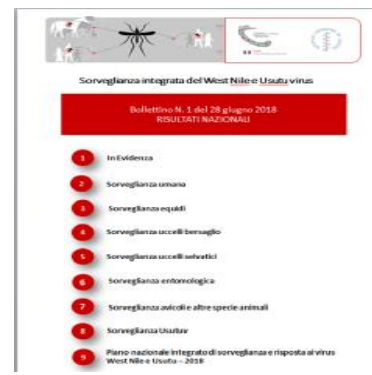
Verify the correctness of notifications in SIMAN

Communicate the outbreak to the Ministry of Health

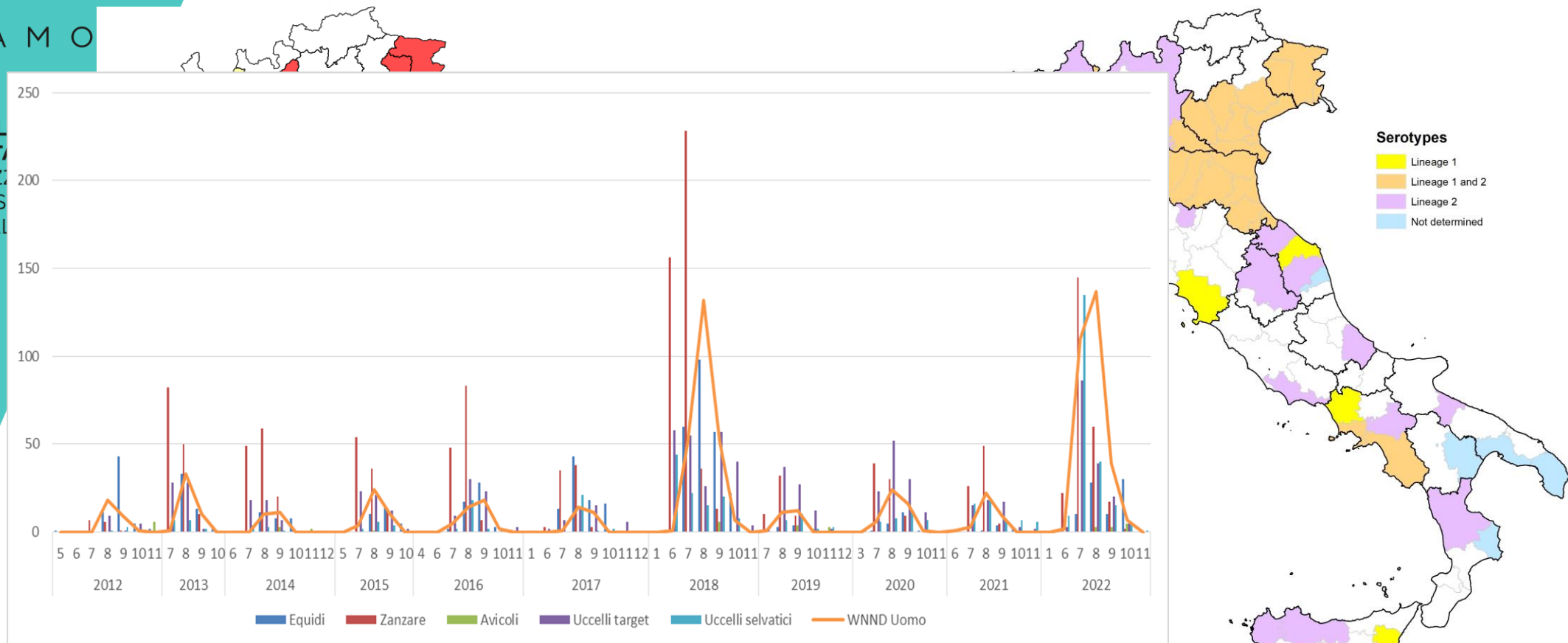
Communicate the outbreak to National Italian blood Centre and National Italian transplant Centre

Report the outbreaks to international bodies

Divulgate the epidemiological situation to general public



WNV numbers: the results of 2024 surveillance



91 wild birds
134 mosquito pools

Updated to 4 September



AIDEO: AI and EO as Innovative Methods for Monitoring West Nile Virus Spread

The recent and massive availability of **Earth Observation (EO)** data and the continuous development of innovative **Artificial Intelligence (AI) methods** can be of great help

- to **automatically identify patterns** in big datasets
- to **make highly accurate predictions**
- to **define intervention priorities** within national diseases surveillance plans

www.aideo.eu

<https://eo4society.esa.int/projects/aideo/>



Article

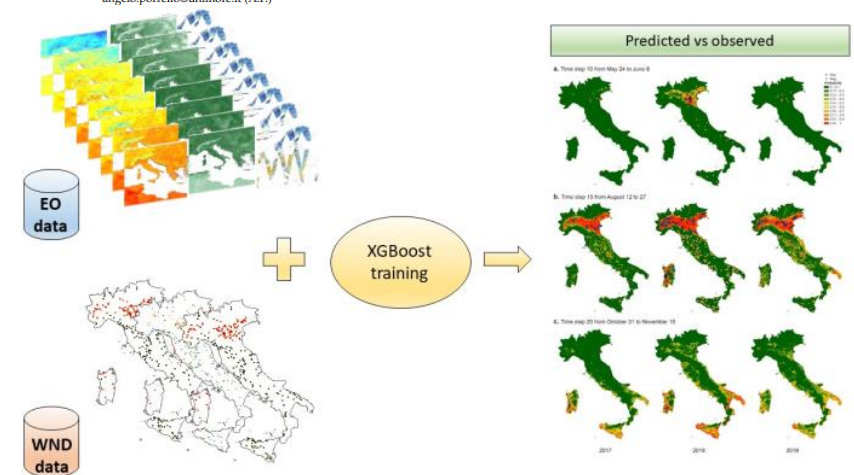
Predicting WNV Circulation in Italy Using Earth Observation Data and Extreme Gradient Boosting Model

Luca Candeloro ^{1,*}, Carla Ippoliti ¹, Federica Iapaolo ¹, Federica Monaco ¹, Daniela Morelli ¹, Roberto Cuccu ², Pietro Fronte ², Simone Calderara ³, Stefano Vincenzi ³, Angelo Porrello ³, Nicola D'Alterio ³, Paolo Calistri ³ and Annamaria Conte ¹

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² Progressive Systems Srl, Frascati, 00044 Rome, Italy; roberto.cuccu@progressivesystems.it (R.C.); pietero.fronte@progressivesystems.it (P.F.)

³ AlmageLab, Engineering Department "Enzo Ferrari", University of Modena and Reggio Emilia, 41121 Modena, Italy; simone.calderara@unimore.it (S.C.); stefano.vincenzi@unimore.it (S.V.); angelo.porrello@unimore.it (A.P.)



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DELL'ABRUZZO
E DEL MOLISE
"G. CAPORALE"



Previsioni

Caricamento previsione
dal 28/08/2024 al 12/09/2024

La previsione utilizza una versione aggiornata (che include i dati WND 2020) del modello descritto [qui](#)

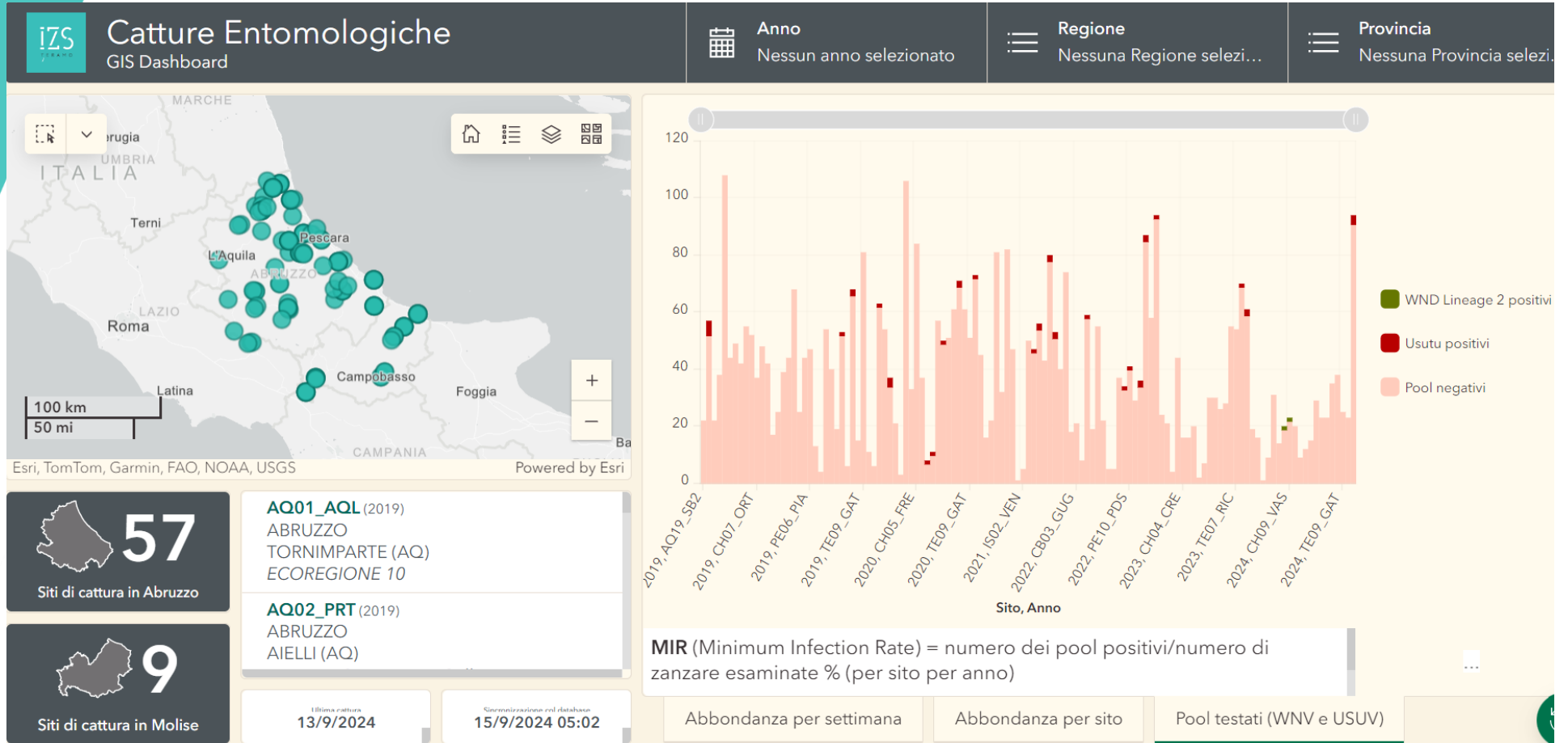
Layer

- Focolai
 - Confermato
 - Sospetto
 - Estinto
- Regioni
- Previsioni
 - 0,0 - 0,1
 - 0,1 - 0,2
 - 0,1 - 0,3
 - 0,3 - 0,4
 - 0,4 - 0,5
 - 0,5 - 0,6
 - 0,6 - 0,7
 - 0,7 - 0,8
 - 0,8 - 0,9
 - 0,9 - 1,0

https://mapserver.izs.it/gis_wn_predictions/#



Mosquitoes dashboard



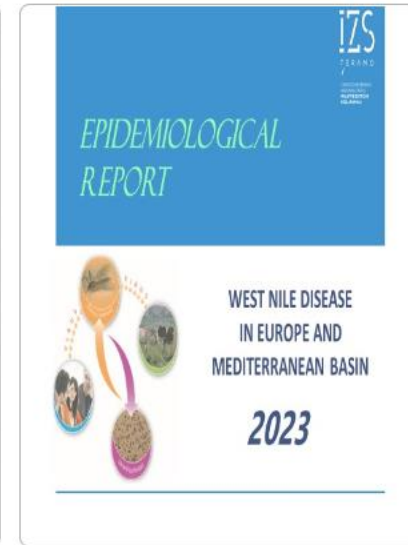
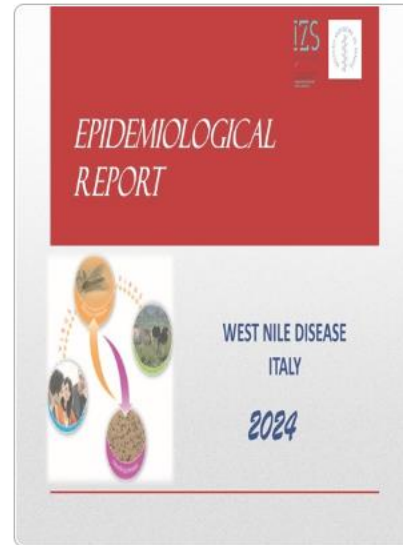
<https://www.arcgis.com/apps/dashboards/282dbe6c42264e9aba37bef5524367b1>

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Reports



https://westnile.izs.it/j6_wnd/home_en

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Raccolta

Integrated surveillance of West Nile and Usutu virus

These annual reports summarize the results of West Nile virus and the Usutu virus surveillance activities in Italy



1 2023



2 2022



3 2021



4 2020



5 2019



6 2018

<https://storymaps.arcgis.com/collections/b50666024702441dac792d0cb3aee32c>

References

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- ✓ Diagne et al. Novel Amplicon-Based Sequencing Approach to West Nile Virus. *Viruses.* 2023 May 27;15(6):1261. doi: 10.3390/v15061261.
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- ✓ Mencattelli et al. Spatial and temporal dynamics of West Nile virus between Africa and Europe. *Nat Commun.* 2023 Oct 13;14(1):6440. doi: 10.1038/s41467-023-42185-7.
- ✓ CDC. Species of dead birds in which West Nile virus has been detected, United States, 1999-2016.
- ✓ https://www.cdc.gov/west-nile-virus/media/files/Bird_species_West_Nile_virus_U.S..pdf



World Organisation
for Animal Health
Founded in 1924

WOAH Regional Workshop on Vector Borne diseases in Asia and the Pacific 19-20 September 2024



Leishmaniasis

Fabrizio Vitale

Direttore Area Biologia Molecolare
Responsabile Centro di Riferenza Nazionale Leishmaniosi (CRENAL)
WOAH Leishmania Reference Lab. Expert
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WOAH Reference Laboratory
for Leishmaniasis

Reference Centre  World Organisation
for Animal Health
Founded as OIE

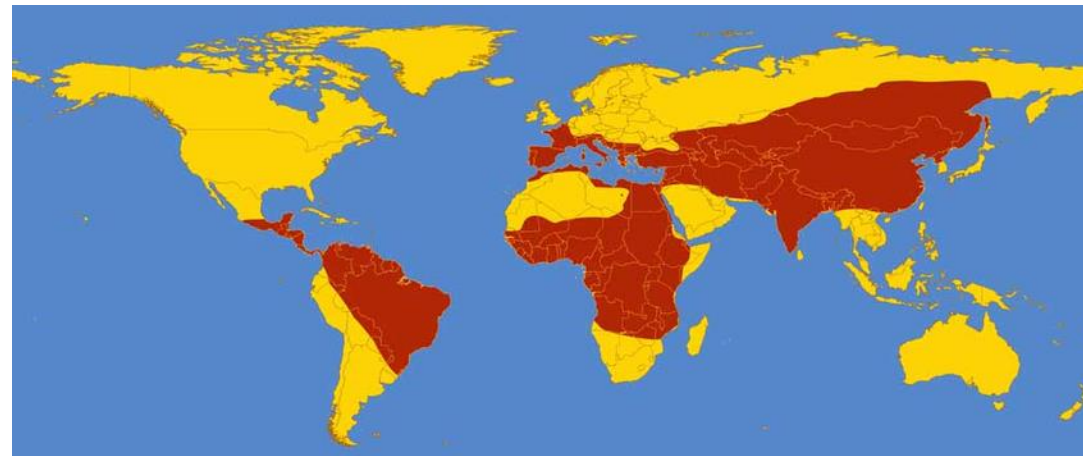


What are we talking about?

The leishmaniases are a group of diseases caused by the protozoa parasite Leishmania.

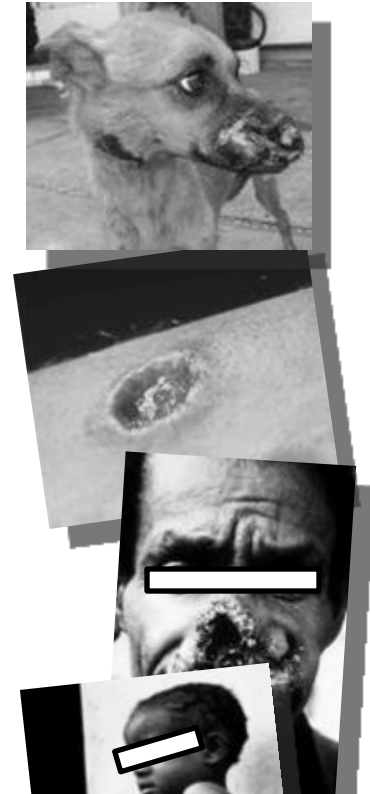
The diseases mainly affects poor people in Africa, Asia and Latin America, and is associated with malnutrition, population displacement, poor housing, weak immune system and lack of resources.

Out of 200 countries and territories reporting to WHO, 99 countries and territories are endemic for leishmaniasis in 2022. This includes 71 countries that are endemic for both VL and CL, 9 countries that are endemic for VL only and 19 countries that are endemic for CL only.



[https://www.researchgate.net/publication/228638177_Trypanosomatids_Odd_Organisms_Devastating_Diseases#:~:text=Trypanosomatids%3A%20Odd%20Organisms,4\(1\)](https://www.researchgate.net/publication/228638177_Trypanosomatids_Odd_Organisms_Devastating_Diseases#:~:text=Trypanosomatids%3A%20Odd%20Organisms,4(1))

But also dogs, cats, humans and many others...



Why dogs, cats and many others?

Of the various Leishmania species most are zoonotic, so a zoonotic disease (zoonosis) is one that "jumps" from an animal to a human. These animals are known as "reservoir hosts" and play an important role in the transmission of Leishmania infection and are able to keep the pathogen that causes the disease, alive, over time, in an ecosystem.

MEMENTO

An excellent reservoir **should**:

- Offers optimal conditions for the reproduction and perpetuation of the microorganism.
- Being keep in touch with humans by sandflies;
- Be receptive to the pathogen;
- Be available for carriers;
- Offer the main food resource for sandflies and share the same habitat.

It is evident that the dog is an example of a good reservoir for Leishmania

But...

Is the dog more likely to be a "victim" rather than a reservoir?



Thus... Why do the programs of mass killing of dogs (conducted in various parts of the world on several occasions, in Brazil) fail to control human visceral leishmaniasis?



Why dogs, cats and many others?

The fatal nature of canine disease suggests that this species is a recent host in evolutionary terms (Campino, 2002).



Table 6. Wild animals reported to be positive for *L. infantum* from Europe, Asia and Africa. Organs or tissues where the parasite was detected are indicated, as well as the techniques employed for detection.

Host	Order	Host	Prevalence	Organs/Tissue Analysed	Methods for Detection	Country	References
<i>Canis aureus</i> (gold)	Carn	<i>Canis lupus</i> (gr)					
<i>Felis silvestris</i> (cat)	Carn	<i>Genetta genetta</i> (coon)					
<i>Herpestes ichnusa</i> (Egyptian mongoose)	Carn	<i>Lutra lutra</i> (European otter)					
<i>Lynx pardinus</i> (lynx)	Carn	<i>Martes foina</i> (beech marten)					
<i>Martes martes</i> (pine marten)	Carn	<i>Meles meles</i> (European wildcat)					
<i>Mustela lutreola</i> (European mink)	Carn	<i>Mustela putorius</i> (European polecat)					
<i>Mustela vison</i> (American mink)	Carn	<i>Panthera tigris</i> (tiger)					
<i>Sciurus vulgaris</i> (red squirrel)	Rodentia	<i>Ursus arctos</i> (brown bear)					
<i>Vulpes vulpes</i> (red fox)	Carn	<i>Pongo pygmaeus</i> (Bornean orangutan)					
<i>Apodemus sylvaticus</i> (wood mouse)	Rodentia	<i>Crociodura russula</i> (white-toothed shrew)	20–50%	blood, BM, liver, skin, spleen	PCR (ITS1) + sequencing, PCR-ELISA (kDNA), qPCR (kDNA) + RFLP + sequencing, PCR (ITS2) + RFLP, smear, culture	Spain	[128,129,159]
<i>Mus musculus</i> (house mouse)	Rodentia	<i>Mus musculus</i> (house mouse)	22–50%	blood, BM, liver, skin, spleen	qPCR (kDNA) + sequencing, PCR (ITS1) + sequencing, PCR-ELISA (kDNA), nPCR (SSU and ITS1) + sequencing, smear	Morocco, Portugal and Spain	[162,163]
<i>Mus spretus</i> (Algerian mouse)	Rodentia	<i>Nesokia indica</i> (short-tailed bandicoot rat)	4.3–42.9%	blood, liver, skin, spleen and serum	qPCR (kDNA), ELISA	Spain	[137,160]
<i>Rattus norvegicus</i> (brown rat)	Rodentia	<i>Rattus norvegicus</i> (brown rat)	5.9–100%	hair, liver, skin, spleen	nPCR (SSU), nPCR (ITS1) + sequencing, qPCR (kDNA), PCR (kDNA), PCR (kDNA) + RFLP, PCR (ITS2) + RFLP, smear	Greece, Morocco,	[129,131,162,
<i>Rattus rattus</i> (black rat)	Rodentia	<i>Rattus rattus</i> (black rat)	7.5–33.3%	blood, BM, liver, skin, spleen	PCR (kDNA) + sequencing, PCR (ITS1) + sequencing, PCR-ELISA (kDNA), nPCR (SSU), nPCR (ITS1) + sequencing, smear, culture, inoculation to hamster, isoenzymes		

BM: bone marrow; Cyt b: cytochrome B; DFA: direct fluorescence antibody assay; GADPH: glyceraldehyde phosphate dehydrogenase; IC: immunochromatography; IF: immunofluorescence assay; ITS: internal transcriber spacer; kDNA: kinetoplast DNA; LN: lymph node; nPCR: nested PCR; qPCR: quantitative PCR; RFLP: restriction fragment length polymorphism; WB: Western blot.



That's all? Vectors (sandflies)

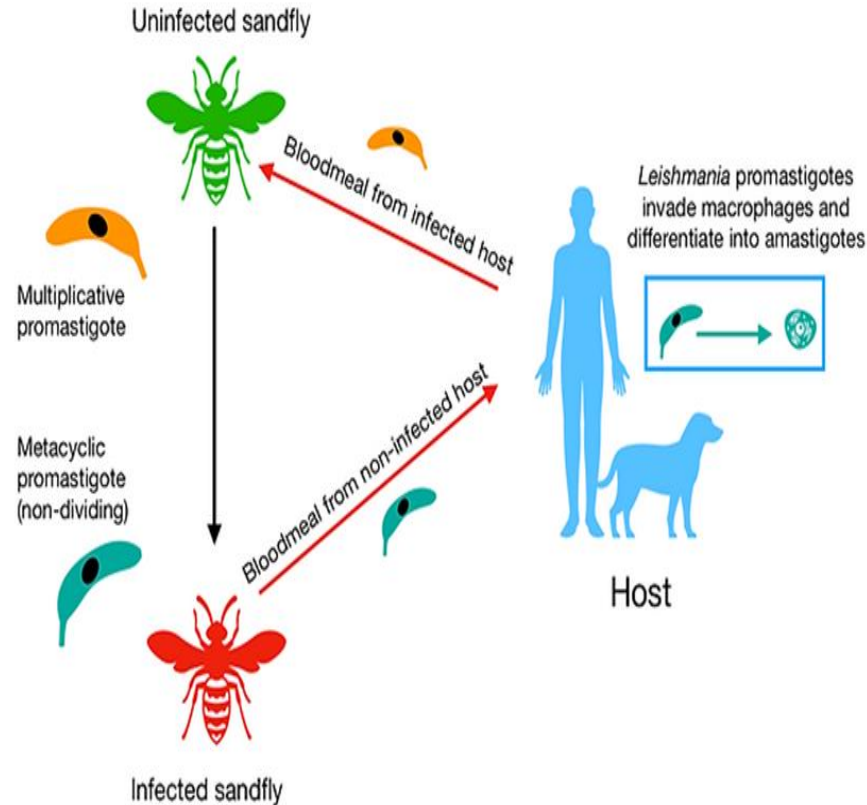


Over 1000 species of sandflies are known but less than 50 are credited with the transmission of Leishmaniasis.

The vector typically:

- She is female
- Performs the blood meal in the evening/night hours
- It has a seasonality (warm-temperate temperatures)
- It remains a few hundred meters from the place of birth

The larvae of sandflies are "terrestrial"



Wild environments

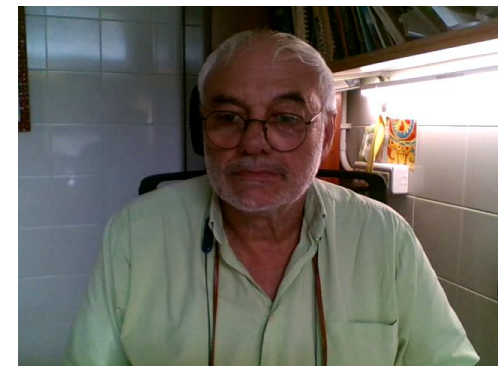
- Ant nests
- Land turtle nests
- Drainage ditches
- Bird nests
- Termite mounds
- Rodent burrows
- Caves
- Shaft recesses
- Forest Floor
- Under and between boulders
- Roots of large trees
- Soil at the base of trees
- Soil under overhanging boulders

Peri-domestic environments

- Animal dens
- Animal shelters
- Chicken coops
- Soil debris and cracks
- Rotting manure heaps
- Earth at the base of old walls
- Under the stones

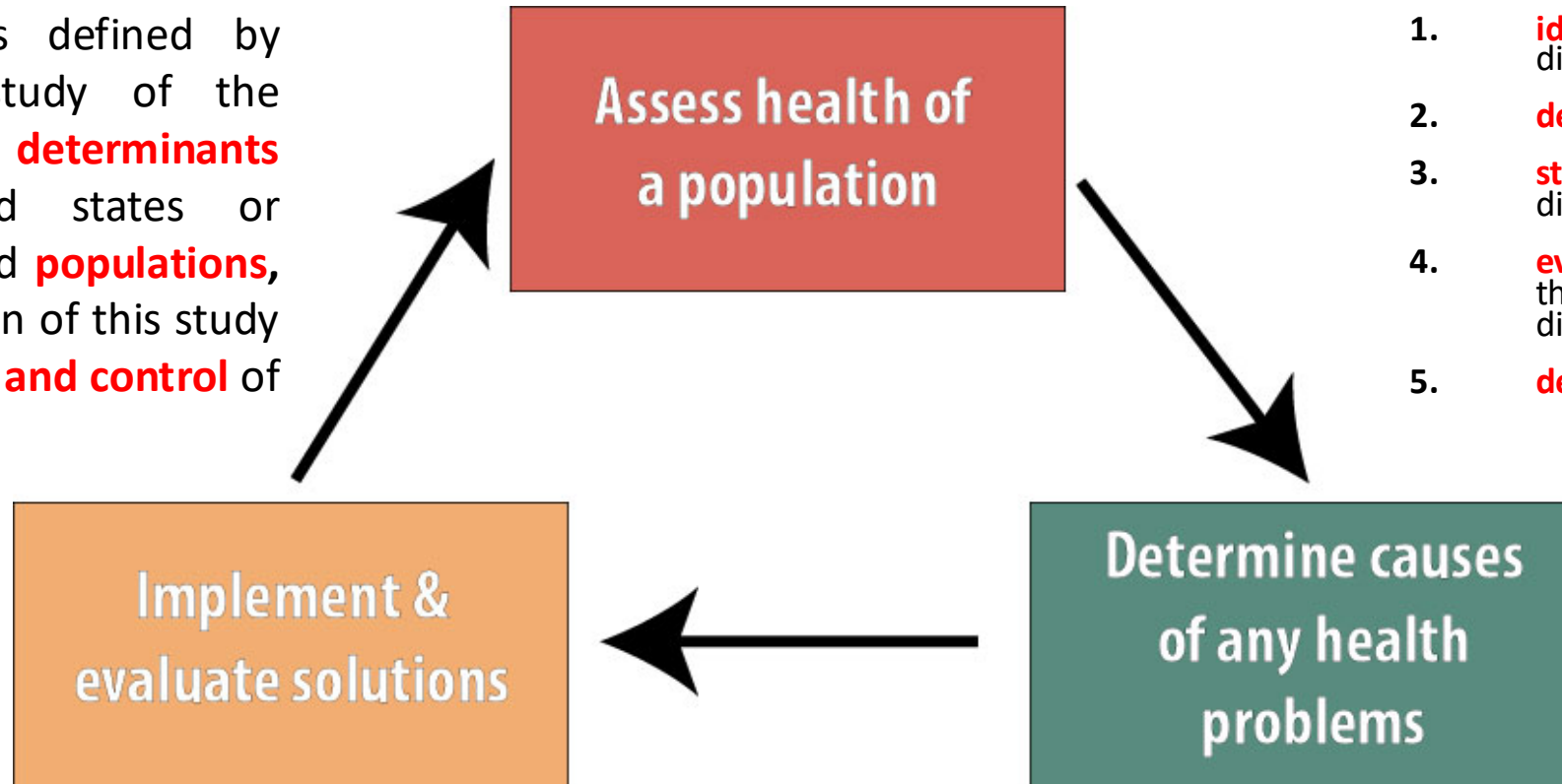
Domestic

- Basements and cellars
- Abandoned houses
- Cracks in floors and walls



Surveillance strategy: focus on Leishmaniasis

Epidemiology, as defined by Last, is “the study of the **distribution** and **determinants** of health-related states or events in specified **populations**, and the application of this study to the **prevention and control** of health problems”.



1. **identify** the etiology or cause of disease.
2. **determine** the extent of disease.
3. **study** the progression of the disease.
4. **evaluate** preventive and therapeutic measures for a disease or condition.
5. **develop** public health policy.

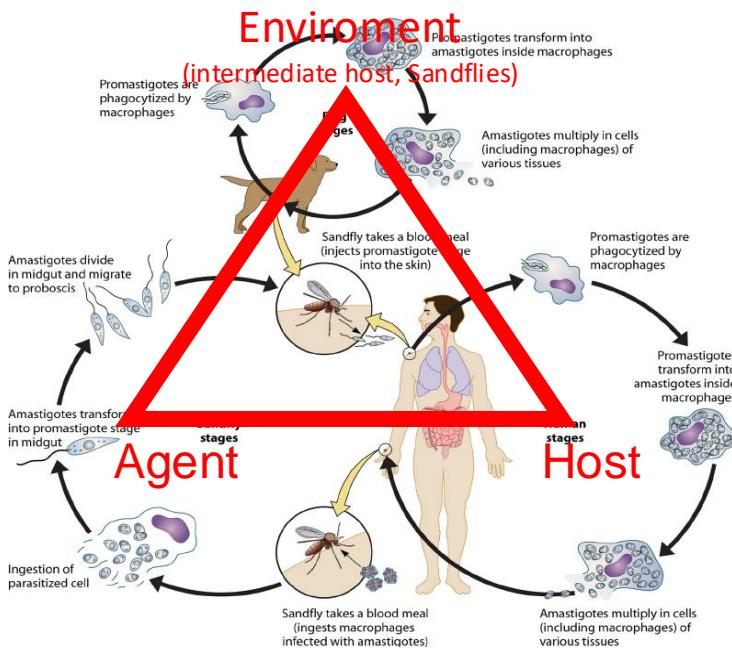




Assess health of a population

1. identify the etiology or cause of disease.

The interaction between living beings, which share the same environment, should be considered as a single dynamic system, in which the health of each component is inevitably interconnected and dependent on the others.



Killick-Kendrick R. *Phlebotomine vectors of the leishmaniasis: a review.* Med Vet Entomol. 1990;4(1):1-24

Distribution range of sandflies according to biogeographical zones

- L. aethiopica*
- L. amazonensis*
- L. arabica*
- L. archibaldi*
- L. aristedes*
- L. (Viannia) braziliensis*
- L. chagasi*
- L. (Viannia) colombiensis*
- L. deanei*
- L. donovani*
- L. enriettii*
- L. equatorensis*
- L. forattinii*
- L. garnhami*
- L. gerbili*
- L. (Viannia) guyanensis*
- L. herreri*
- L. hertigi*
- L. infantum*
- L. killicki*
- L. (Viannia) lainsoni*
- L. major*
- L. mexicana*
- L. (Viannia) naiffi*
- L. (Viannia) panamensis*
- L. (Viannia) peruviana*
- L. (Viannia) pifanoi*
- L. (Viannia) shawi*
- L. tarentolae*
- L. tropica*
- L. turanica*
- L. venezuelensis*

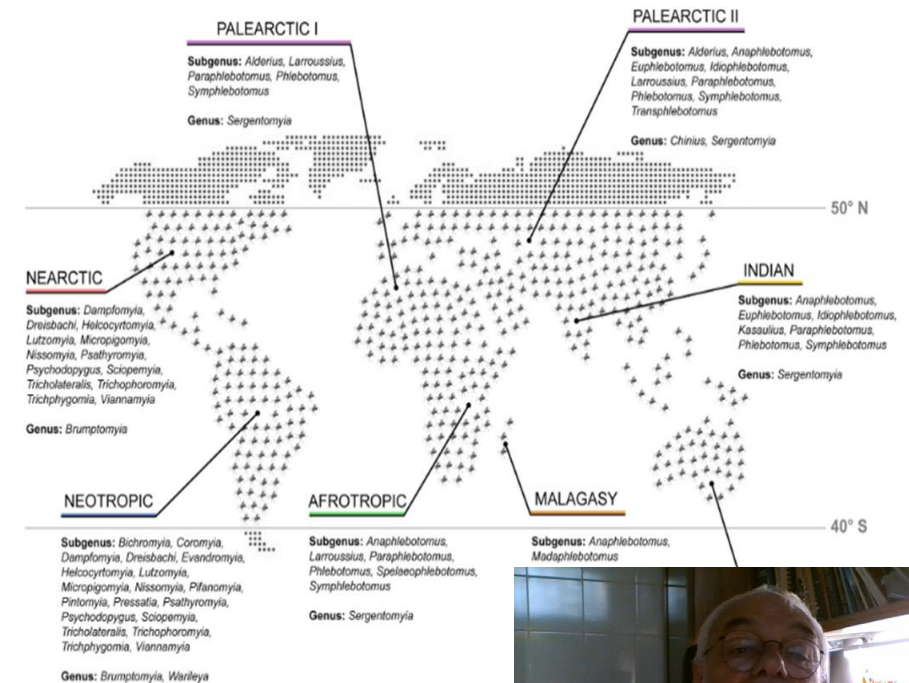


Fig. 1 Sand fly distribution map by genera/subspecies. Sand flies have a global distribution (excluding New Zealand and the Pacific islands). In the map, the classification based on a conservative approach are listed based on their presence in Neotropical (dark blue), Afrotropical (green), Malagasy (orange), Australia (light blue)

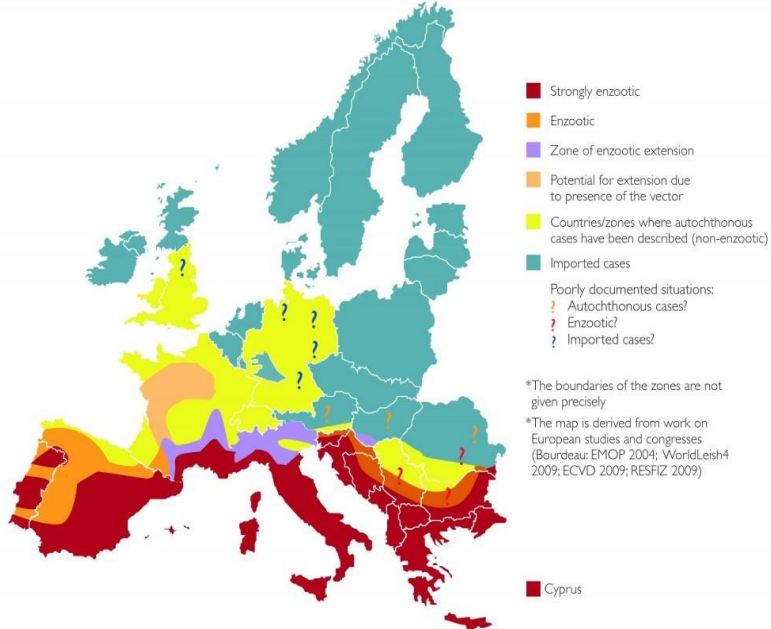




Assess health of a population

Canine leishmaniosis in Europe (Pr. P. BOURDEAU Original)

(Courtesy Prof P. Bourdeau) shows the current broad epidemiological situation in Europe.

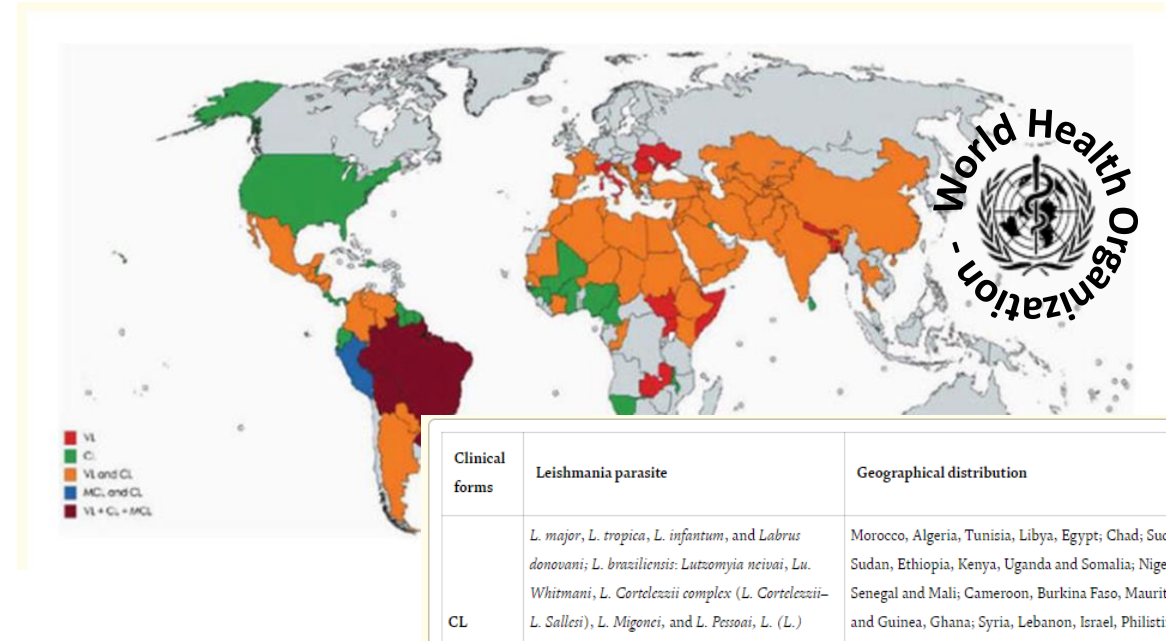


- Canine leishmaniosis is present in >70 countries
- Infected dogs in Italy, Spain, France and Portugal are around 2.5 million
- Many more in South America

1. **determine** the extent of disease.

The surveillance activity is focused in Europe on the main synanthropic reservoir of the disease: the dog and develops at a regional level on the basis of some considerations:

- Canine reservoir on the rise in non-endemic areas
- Rapidly spreading vector phlebotomine habitat
- Interregional Dog Handling
- Improved information flow Human and animal health



OPEN ACCESS PEER-REVIEWED CHAPTER
Epidemiology of Leishmaniasis
Stephen Kyari

Clinical forms	Leishmania parasite	Geographical distribution
CL	<i>L. major</i> , <i>L. tropica</i> , <i>L. infantum</i> , and <i>Labrus donovani</i> ; <i>L. braziliensis</i> ; <i>Lutzomyia neivai</i> , <i>Lu. Whitmani</i> , <i>L. Cortelezzi</i> complex (<i>L. Cortelezzi</i> - <i>L. Sallesi</i>), <i>L. Mignonci</i> , and <i>L. Pessoi</i> , <i>L. (L.) peruviana</i> , <i>L. lainsoni</i> , <i>L. amazonensis</i> , <i>L. infantum chagasi</i> , <i>L. Mexicana</i> , <i>Leptodactylus colombiensis</i> and <i>L. equatoriensis</i>	Morocco, Algeria, Tunisia, Libya, Egypt; Chad; Sudan, South Sudan, Ethiopia, Kenya, Uganda and Somalia; Niger, Nigeria, Senegal and Mali; Cameroon, Burkina Faso, Mauritania, Gambia and Guinea, Ghana; Syria, Lebanon, Israel, Philistines, Iran, Saudi Arabia, Iraq, Pakistan, Afghanistan, Turkmenistan; Greece; United State of America; Bolivia, Brazil, Colombia, Peru; Sri Lanka, China, Nepal Bangladesh; India, and
MCL	<i>L. viannia braziliensis</i> , <i>L. viannia amazonensis</i> , <i>L. viannia panamensis</i> and <i>L. viannia guyanensis</i>	
DCL	<i>L. Mexicana</i> , <i>L. aethiopicus</i> , <i>L. amazonensis</i> , <i>L. pifanoi</i> , <i>L. guyanensis</i> , and <i>L. panamensis</i>	
VL	<i>L. donovani</i> , <i>L. infantum</i> , <i>L. tropica</i> , <i>L. chagasi</i>	

Table 2.
The table below shows the clinical form of leishm





Determine causes of any health problems

1. study the progression of the disease.



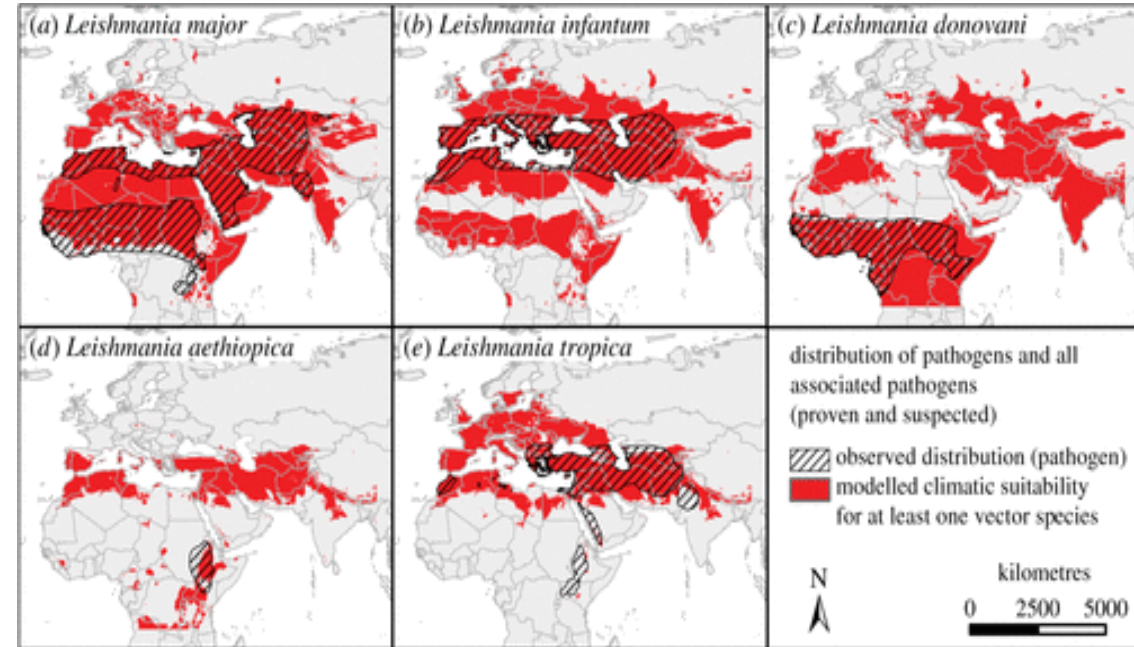
Where are we?

Its strategic location at the centre of the Mediterranean has made the island a crossroads of history, a pawn of conquest and empire, and a melting pot for ethnic groups, vegetal inflorescence and...arthropods!

Why do we care so much about climate changes?

The arthropod vectors of diseases are **ETEROTHERMIC** organisms (without temperature regulation system) and therefore they depend on the environmental temperature

Climatic suitability of vector species and distribution of associated pathogens. **Hatched areas** indicate the observed distribution of the five Leishmania species, **red** indicates areas with suitable climatic conditions based on the binary modelling results using equal training sensitivity and specificity threshold projected for at least one of the associated vector species (with confirmed or strongly suspected vector-competence for the respective Leishmania species).



Leishmaniasis in Eurasia and Africa: geographical distribution of pathogens and associated vector species. *PLoS ONE* 2014; 9(10): e109834. DOI: (10.1098/rsos.190334)



Determine causes of any health problems

1. **evaluate** preventive and therapeutic measures for a disease

Preventing Leishmaniasis

KEY POINTS

- There are no vaccines or drugs to prevent leishmaniasis infection.
- The best way people can prevent infection is to protect themselves against sand fly bites.



Joint Effort – Applied Surveillance
Purpose:

- ✓ Track sand fly populations
- ✓ Evaluate Control effectiveness
- ✓ ID sand fly & disease “hot spots”
- ✓ Determine Leishmania infection rates
- ✓ Which trap designs are most effective

Prevention

Leishmaniasis is mainly prevented by suppressing and killing sandfly vectors which transmit the disease ([vector control measures](#)). Vector control measures are critical to reducing disease incidence [in the absence of an effective vaccine](#).

The primary strategies of vector control are [indoor residual spraying](#) (IRS) and [long-lasting insecticide-treated nets](#) (LLINs) with [pyrethroid insecticides](#). IRS involves spraying the walls and other surfaces of houses and buildings, LLINs offer protection to individuals.

Table 5 Current treatment protocols for canine leishmaniasis [27]

Drugs	Dosages	Main side effects	References
Meglumine antimoniate*	75-100 mg/kg once a day or 40-75 mg/kg twice a day for 4 weeks, S.C.**	Potential nephrotoxicity Cutaneous abscesses/cellulitis	[52,55,57,81-83]
Miltefosine*	2 mg/kg/once a day for 28 days P.O.	Vomiting Diarrhea	[83-85]
Allopurinol	10 mg/kg twice a day for at least 6-12 months P.O.	Xanthine urolithiasis	[51,59,86-89]

*Registered for veterinary use in most European countries; both drugs are commonly recommended in combination with allopurinol.

P.O.: per os; S.C.: subcutaneous

**Treatment prolongation by 2-3 weeks may be considered if patient improvement is insufficient.

Table 6 Treatment of canine leishmaniasis - recommended monitoring of clinicopathological parameters and serology including frequency of follow up [27]

Parameters	Frequency
Clinical history and complete physical examination	After the first month of treatment and then every 3-4 months during the first year. Later on, if the dog is fully recovered clinically with treatment, a recheck would be recommended every 6 months or once a year.
Routine laboratory tests: Complete CBC, biochemical profile, serum electrophoresis (optional) and complete urinalysis including UPC in proteinuric dogs.	
Serology*	Not before 6 months after initial treatment and every 6 months or once a year thereafter.
Real time PCR	Can optionally be carried out. The usefulness of this assay is undetermined.

*Some dogs present a significant decrease in antibody levels (more than a two-fold dilutions difference) with clinical improvement within 6 months to 1 year of treatment. Other dogs might not have a decrease in antibody levels. In contrast, a marked increase of antibody levels (more than two-fold elevation between monitoring points) is observed especially in dogs following the discontinuation of treatment [27].



Solano-Garcia
<http://www.woah.org>
LeishVet
of canine



Implement &
evaluate solutions

1. **develop** public health policy.



What needs

- *Public health surveillance at the International level* (mandatory notification system in all countries?)
- Educate people on interventions against sandflies
 - Insect repellents Insecticides
 - Use of insecticide impregnated nets and bed nets
 - Dog: topical applications and impregnated collars
- Further research on
 - Alternative modes of transmission
 - Effective vaccine for human leishmaniasis
 - immunisation strategy for human populations
 - Effective vaccine for canine leishmaniasis
 - to control the infections in reservoir population (dogs, cats, hares,
 - Better predictive modelli

Actually

Sandfly biting activity is strongly seasonal, restricted to summer months in most areas (e.g. Southern Europe, below 800m above sea level)

Future & climate change

Prolonged activity periods and shorter diapause periods (overwintering)
Extend northwards and into higher altitudes

At present, no good prediction models available

*Leishmaniasis remain **widespread and under-reported**. Measures for **leishmaniasis prevention and control**, access to valid diagnostic methods and guidelines, and access to effective treatments **vary considerably between countries**. This variation and the lack of resources in some countries or regions, could have important disease implications including increased incidence; unnoticed spread of *Leishmania spp.* into new areas; increased treatment failure and development of resistance to treatments.*

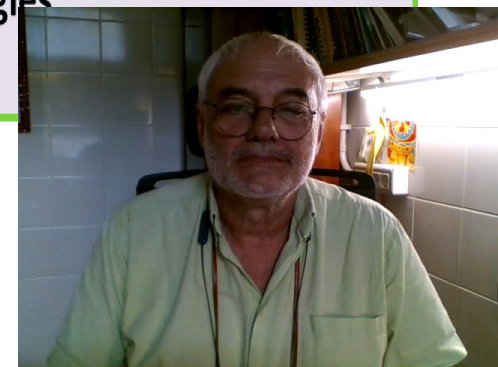




Leishmaniasis: An example of how essential the concept of One Health is



- **Medical doctors** to recognize and treat human clinical forms (CL / VL)
- **Veterinarians** to recognize and treat animal clinical forms
- **Biologists** to optimize research laboratories
- **Entomologists** to monitor vectors
- **Wild experts** to identify non-anthropized reservoirs
- **Ecologists** to make health authorities responsible for the principles of public hygiene
- **Epidemiologists** to outline surveillance strategies
- **Media** to inform about risks and prevention



Thank you ...

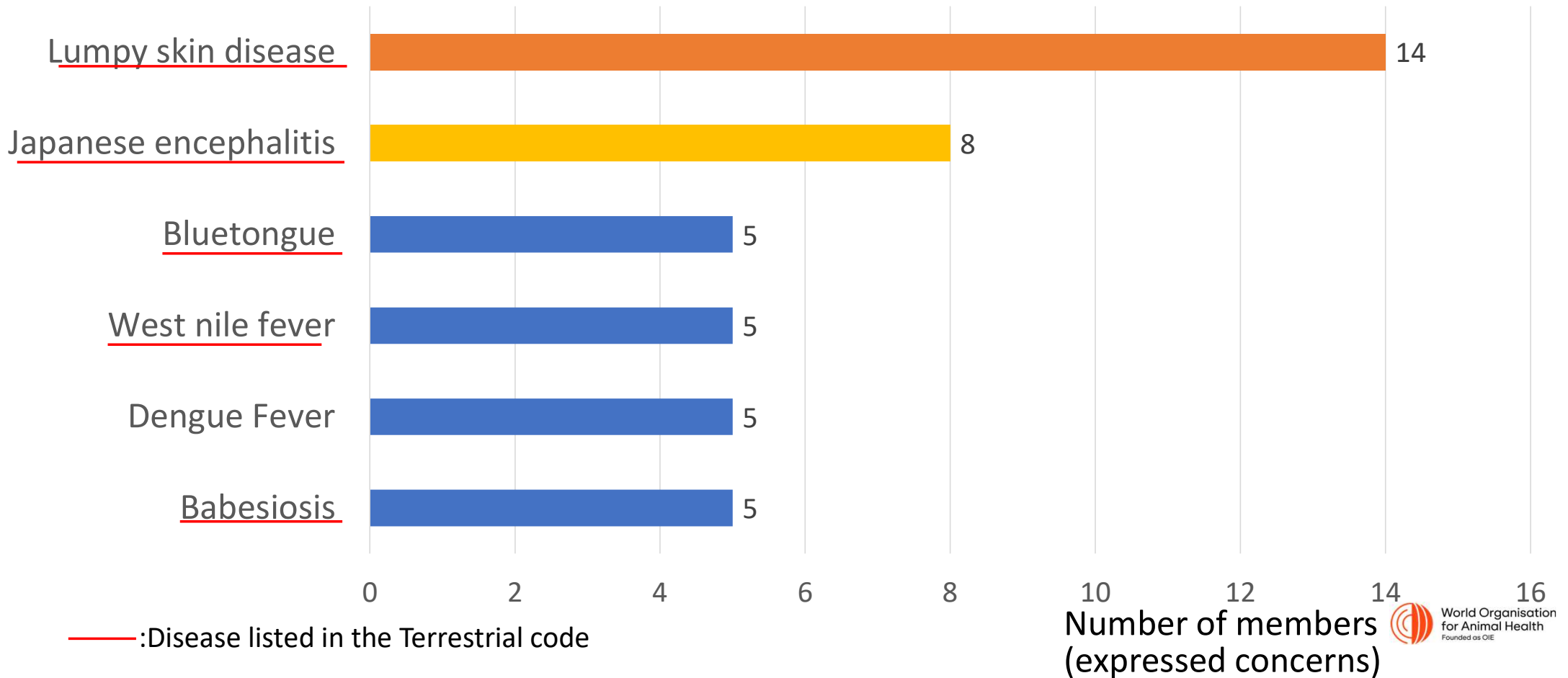
Summary from pre-meeting questionnaire

Arjun Pandit, Hokkaido University
Kevin Chyi, D.V.M, APHIA Chinese Taipei
WOAH Interns

19 September 2024
Tokyo, Japan

VBD(s) concerned by Regional Members-1/2

Diseases of concerns



VBD(s) concerned by Regional Members-2/2

LSD, JE, BTV, WNF, Dengue fever, Babesiosis are of the most concerns,

1. Widespread in the region
2. Doing their best to prevent, eradicate or contain in the territory
3. Zoonotic diseases

<u>Anaplasmosis</u>	<u>Epizootic hemorrhagic disease</u>	Severe Fever with Thrombocytopenia Syndrome(SFTS)
<u>Theileriosis</u>	<u>Q fever</u>	Hepatozoon
<u>African horse sickness</u>	Akabane disease	Chuzan virus
<u>African swine fever</u>	Crimean-Congo haemorrhagic fever	Chikungunya
<u>Trypanosomiasis</u>	Malaria*	Bovine ephemeral fever
<u>Leishmaniosis</u>	<i>Mycoplasma haemocanis</i>	<i>Ehrlichia canis</i>
<u>Rift valley fever</u>	Dirofilaria	<i>Rickettsia felis</i>
	Schistosomiasis	

—: Diseases listed in the Terrestrial code

Responses to VBD(s)-1/2

- **Surveillance**
 - Vector surveillance (mosquito, biting midge, stable fly, fly, tick...)
 - Active surveillance (LSD, JE, BTV, WNF, AHS, Akabane disease, RVF ...)
- **Preventive measures**
 - Vector control (Repellant, insecticide, vector trap...)
 - Biosecurity on farm
 - Vaccination (LSD, JE, Q-fever, Akabane disease...)
 - Contingency plan
- **Response during a disease outbreak**
 - Movement control (LSD, JE, AHS, ASF...)
 - Treatment (Q-fever, Surra, tick...)
 - Stamping out (LSD, AHS, ASF...)

Responses to VBD(s)-2/2

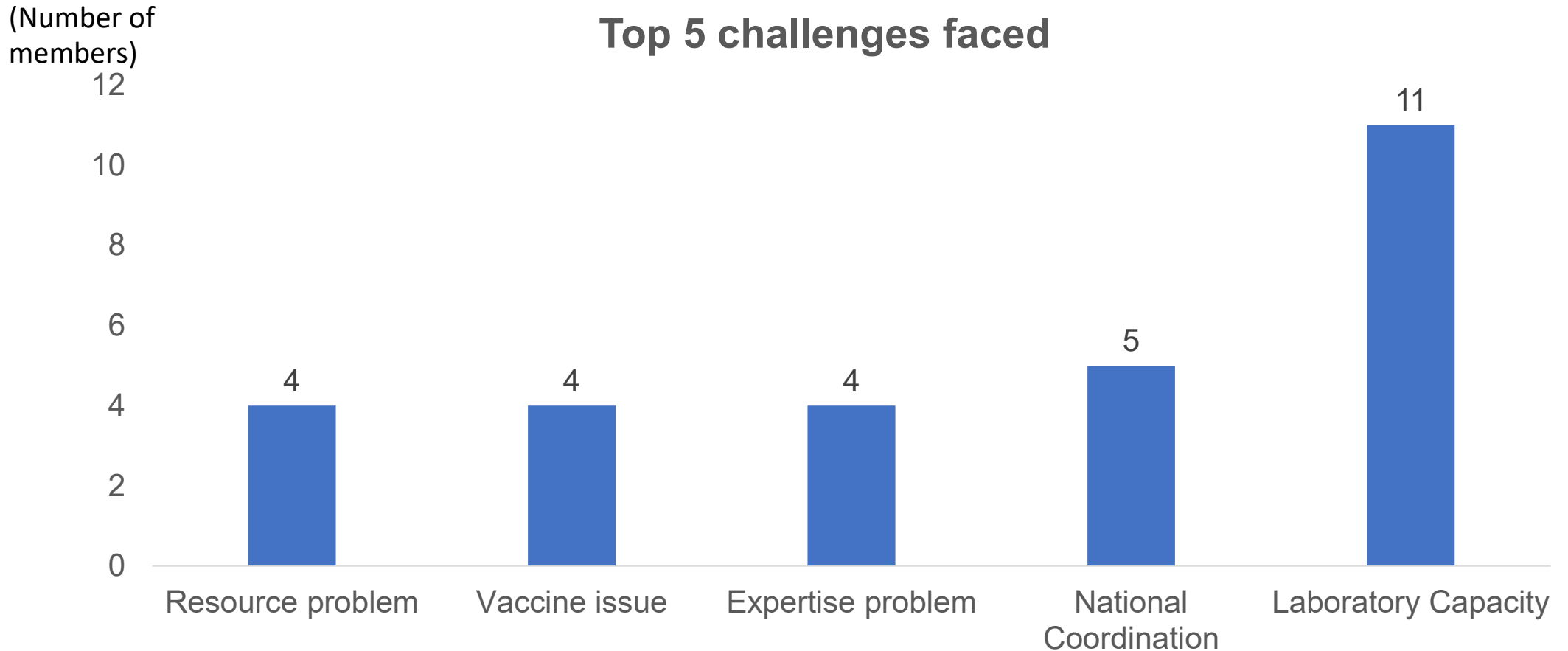
- **Public awareness**
 - Training and education
 - Communication (Using One Health approach. Conducting awareness campaigns to the general public)
- **Diagnostic ability**
 - Development of the detection method (diagnostic kit for detection Surra)
 - Diagnostic training

Impact of actions

- **Successfully prevent or eradicate VBD (14)**
 - Reduce the risk of exposure (11)
 - To regain VBD-free status (3)
- **Diagnostic and Surveillance efficiency increased (8)**
 - Enhanced diagnostic and surveillance ability (6)
 - Importance of early warning system (2)
- **Vaccination is effective in controlling VBD (5)**
 - Vaccination is an effective measure to combating VBD (4)
 - Make stockpiles of vaccines possible (1)
- **Other impact**
 - Strengthen awareness and education, data-sharing
 - Improved policy and strategies, established list of priority, identify the risk, enhanced biosecurity

Summary of challenges faced by members

Top 5 challenges faced



Other problems: Awareness problems, under-reporting, and surveillance problems

Possible solutions to overcome the challenges

- **Capacity development**

- Diagnostic to field staff
- Continued education
- Vector and vaccine development training

- **Coordination with partners**

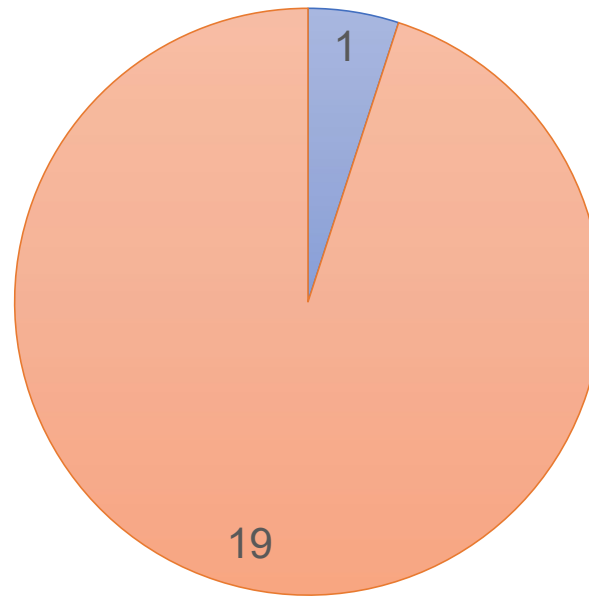
- Strengthen cross-border collaboration between partners
- Cost-sharing (PPP with development partners),
- Coordination from sub-national to International level

- **Surveillance facilitation**

- **Data sharing and some innovations** (PCR on feral pig tonsils, chew ropes for monitoring tools for pigs: Australia),
- Country-level sustainable **active surveillance**

Collaboration with other sectors under One Health approach

OH status of members



■ No OH experiences ■ Having experience of OH collaboration

Collaboration with other sectors under One Health approach

- **Multisectoral collaboration**

- Shared surveillance information/Protocol/Publication/collaborative risk assessments (New Zealand)
- Multisectoral collaboration from the sub-national to the international level
- Co-writing of contingency plans (New Caledonia)

- **Joint surveillance**

- Joint surveillance of feral animals and other animals (Australia)
- Arbovirus surveillance (Japan)
- JE and wildlife disease surveillance (Malaysia)
- integrated surveillance (New Caledonia)

Possible actions to strengthen One Health approach

- **Multisectoral collaboration**
 - Data and Information Sharing
 - Joint surveillance framework
 - Multisectoral expert networking
 - Joint research projects
 - Collaborative preparedness resilience
 - Zoonoses simulation exercises
 - Joint capacity building
- **Regular stakeholder meeting**
 - Frequent meetings between stakeholders
 - Creating platforms for knowledge-sharing
 - Information sharing frameworks
 - Regular resource funding.

Expectations for the VBDs workshop

- **Experiences of members**

- VBDs epidemiology in the climate change context
- Success stories and experience of members (Epizootic hemorrhagic diseases in Europe, Akabane disease and Blue Tongue in China and India)
- SFTS management
- Early warning systems
- Contingency plans of different members

- **Information from experts on specific diseases**

- Management strategy
- Latest global epidemiology of LSD, WNF, and AHS
- Cost-effective early warning system of VBDs.

Thank you

Zoonotic and human related diseases and their control

Ken Maeda, Ph.D., D.V.M.

Director

Department of Veterinary Science

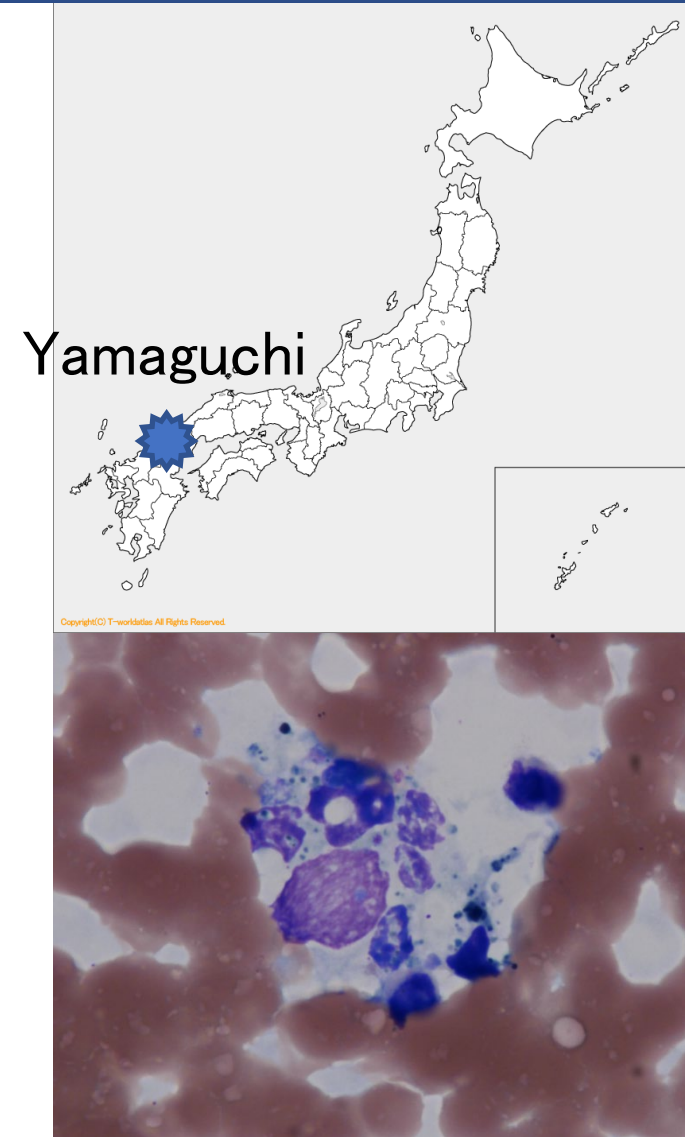
National Institute of Infectious Diseases (NIID)

Major emerging viral zoonosis in Japan

Date	Emerging viral diseases	Vector	Animals
Dec, 2012	First report of severe fever with thrombocytopenia syndrome (SFTS) in Japan	Tick	Many mammals
Aug, 2014	Endemic of dengue fever in Tokyo after 69 years	Mosquito	Non-human primates (sylvatic type)
Jul, 2016	Tick-borne encephalitis after 23 years	Tick	Rodents, Wild animals
May, 2019	First discovery of Yezo virus in the world	Tick	Wild animals?
Nov, 2019	First report of B virus infection in Japan (2 cases)		Macaque
Jan, 2020	Pandemic of COVID-19 in Japan		Bat?
May, 2020	Rabies in Japan after 14 years		Dog, Bats
Jul, 2022	Mpox in Japan		Rodents etc.
Summer, 2022	Oz virus infection in Chiba	Tick	Wild animals

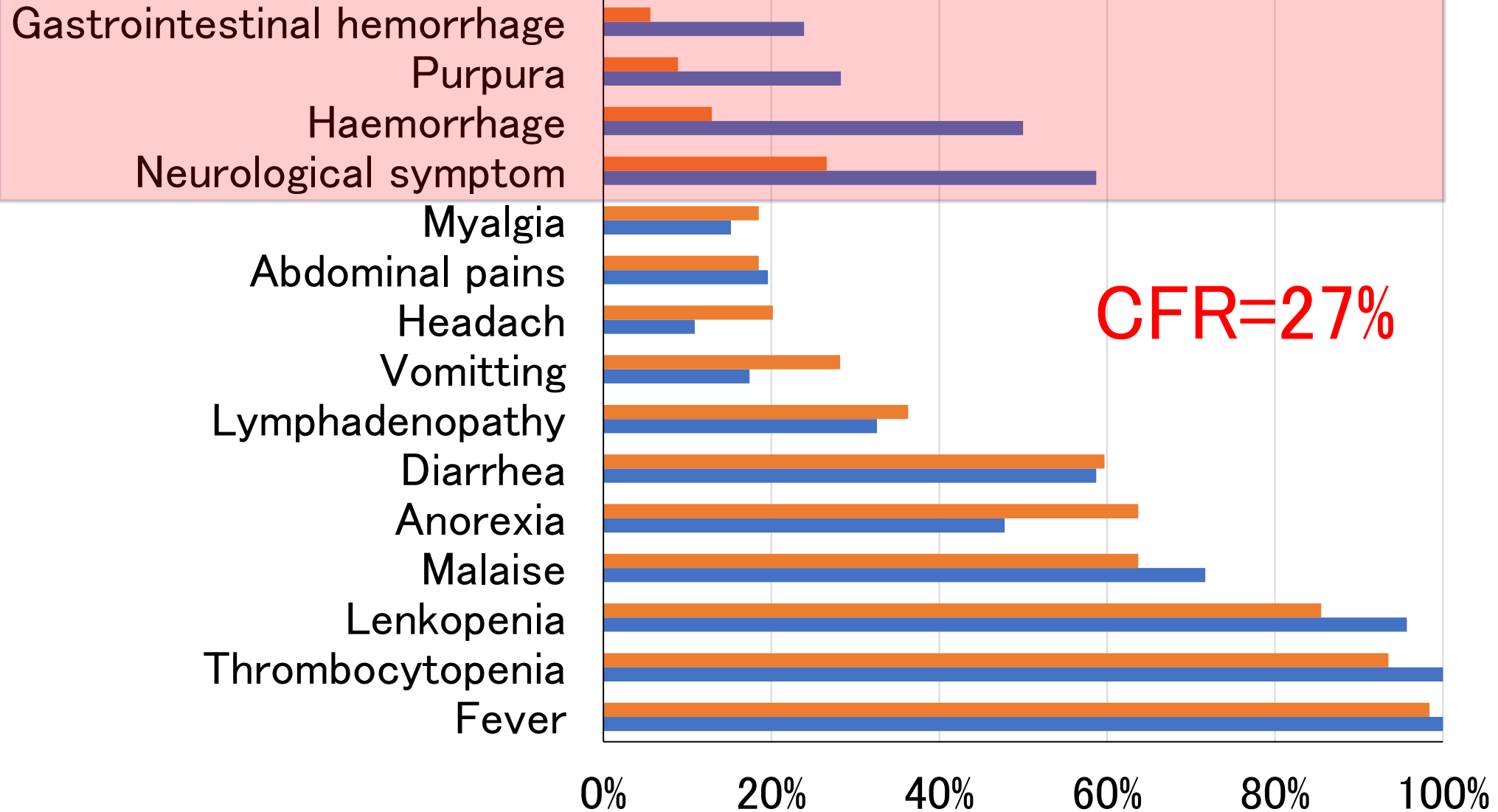
The first case of SFTS in Japan

- Patient: Female aged 50's without overseas travel
- Onset time and location: Autumn 2012, Yamaguchi prefecture
- Symptoms: Fever, general fatigue, vomiting, melena (**bloody diarrhea**)
- Laboratory findings: Leukopenia (**400** /mm³) and Thrombocytopenia (**8.9X10⁴** /mm³) in TBC;
Elevated AST, ALT, LDH, CK;
Extremely elevated ferritin in serum chemistry;
Hematuria and proteinuria in urinalysis;
Hemophagocytosis with hypocellularity findings in bone marrow aspiration



Clinical symptoms in human patients

Recovery Death



Distribution and age of SFTS patients in Japan

SFTS patients

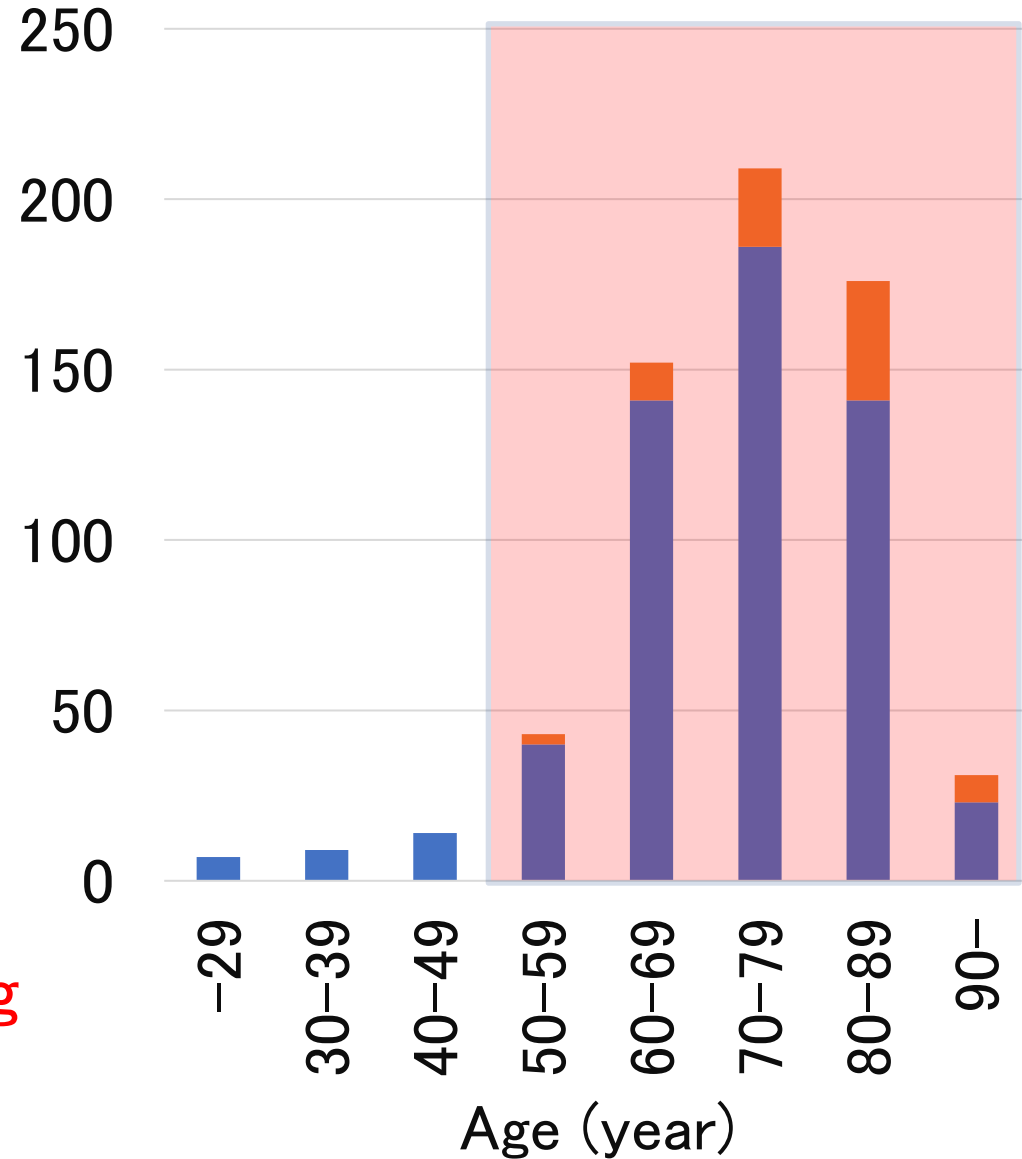
0

83

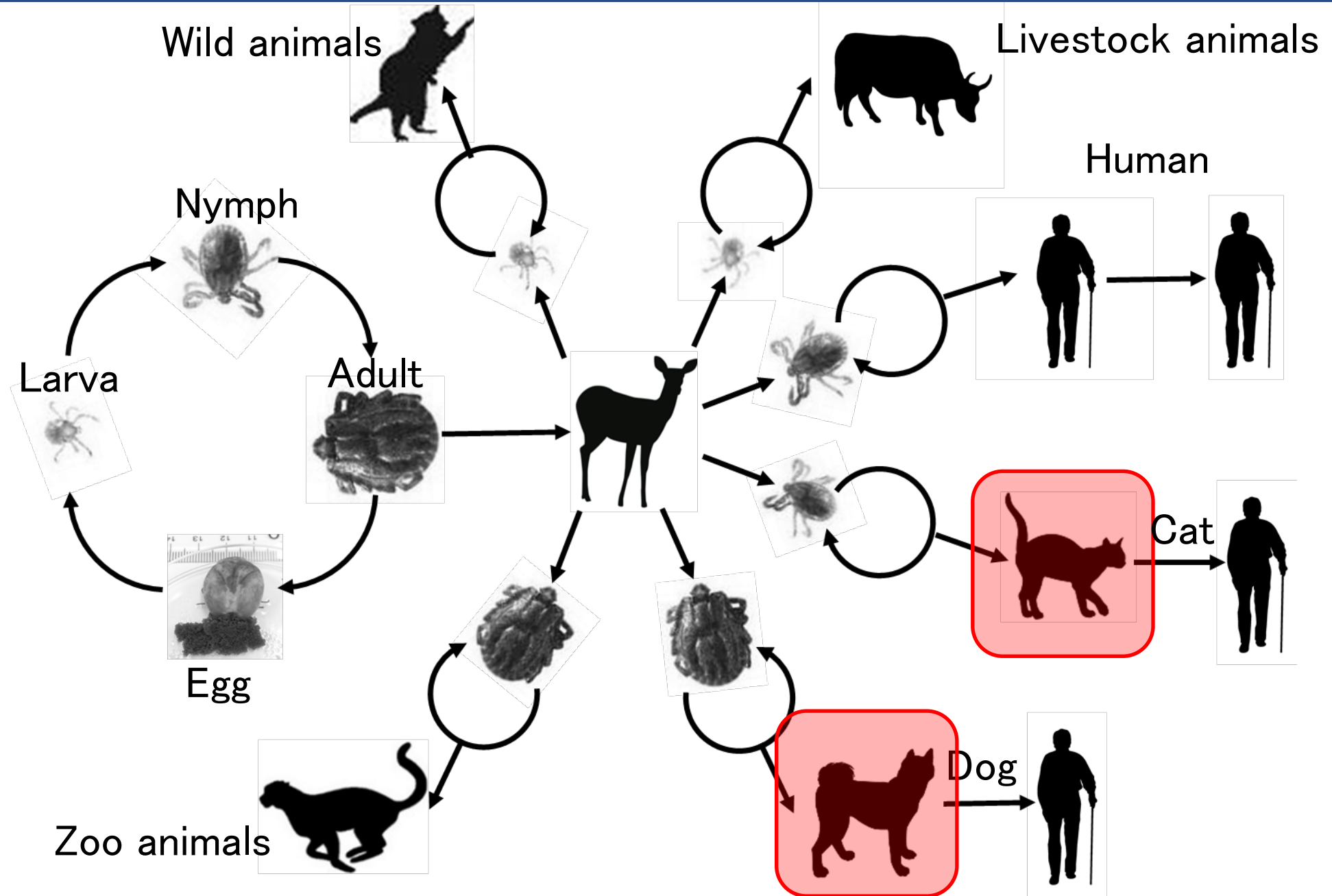
■ Recovery ■ Death



SFTSV is expanding to the east



Transmission cycle of SFTSV



Clinical symptoms in SFTS cats



No. of Positive
/No. of Total

Ratio

Low activity ▪ Anorexia

97/97

100%

Vomiting

44/95

46%

Diarrhea

5/95

5%

Jaundice

43/43

100%

Death

41/69

59%

First SFTS dog and symptoms in SFTS dogs



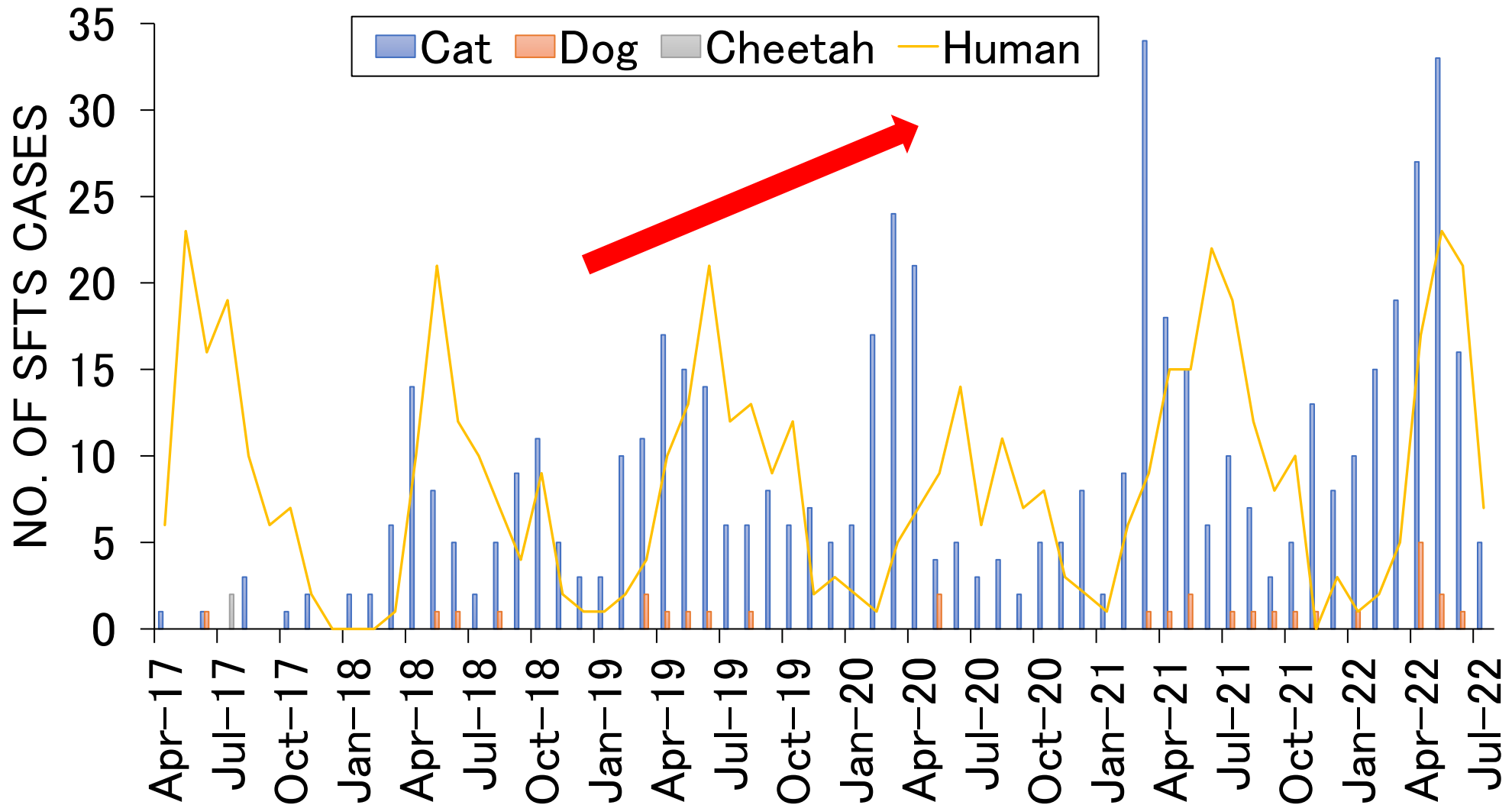
- June, 2017
- Mix
- Spayed female
- 4 years old
- Every year 6 combination vaccine and rabies vaccine
- Heartworm and tick preventives

Clinical findings in seven SFTS dogs

Clinical findings	Number of dogs with abnormal values (%)
Anorexia and less activity	7 (100%)
Vomit	2 (29%)
Loose stool	2 (29%)
Dead	3 (43%)
Fever ($>39^{\circ}\text{C}$)	7 (100%)
Leukocytopenia ($<6,000/\mu\text{l}$)	7 (100%)
Thrombocytopenia ($<200,000/\mu\text{l}$)	7 (100%)
Tick parasite	5 (71%)

Owner of this SFTS dog was also infected with SFTSV without tick-bite

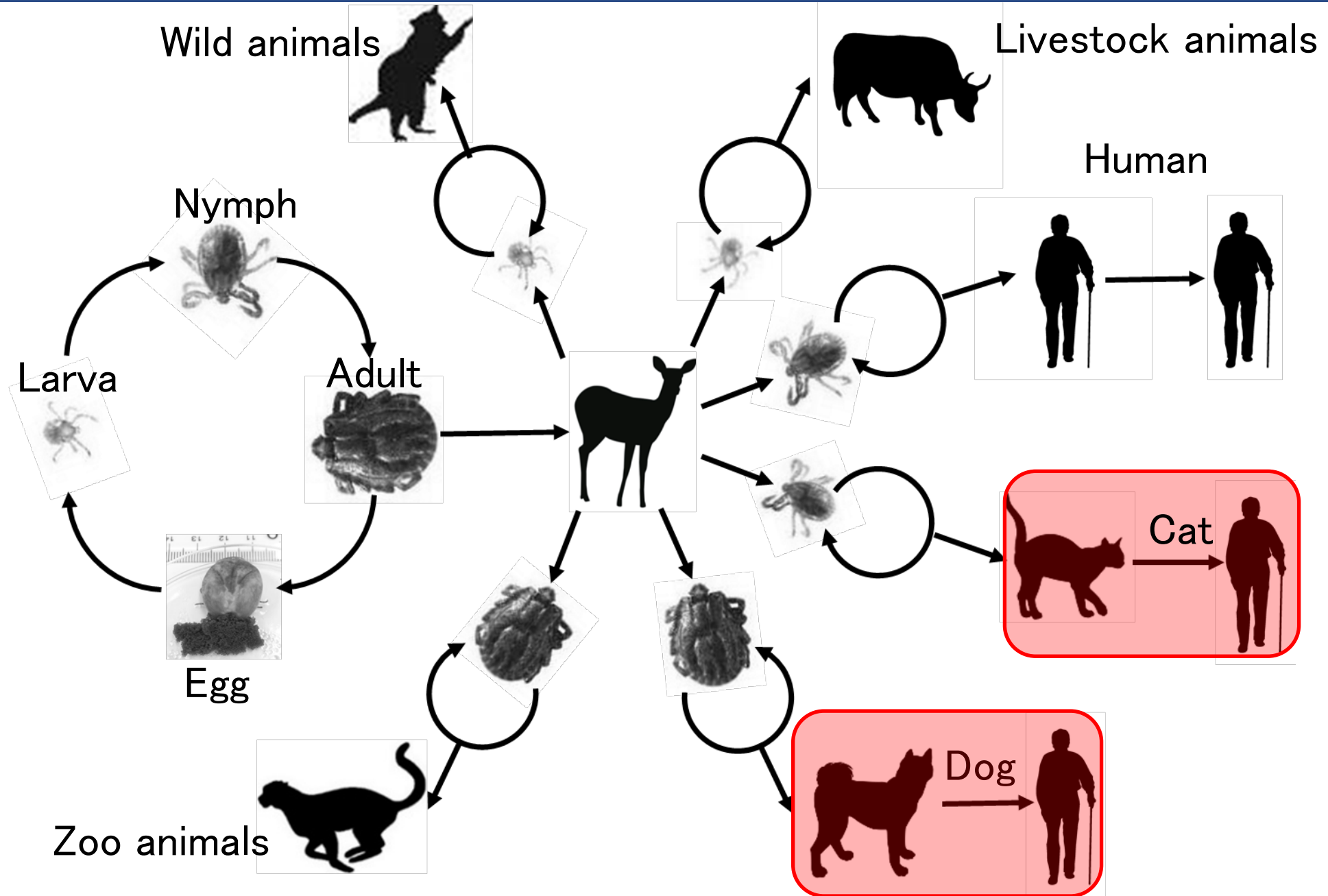
Number of SFTS cases in animals and humans



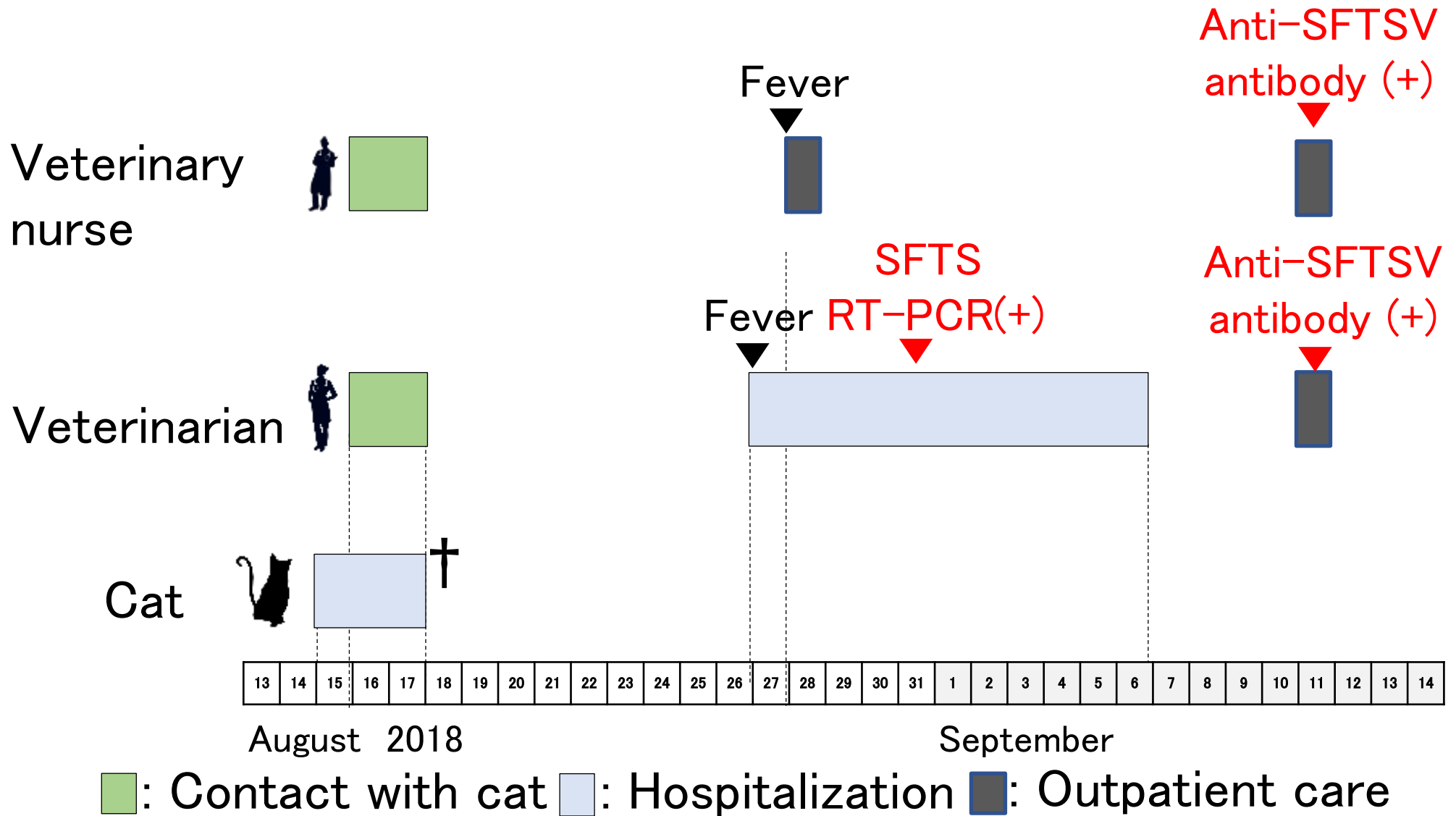
The tendency is similar to that of human cases.

Cat cases are more than human cases

Transmission from cats and dogs to human



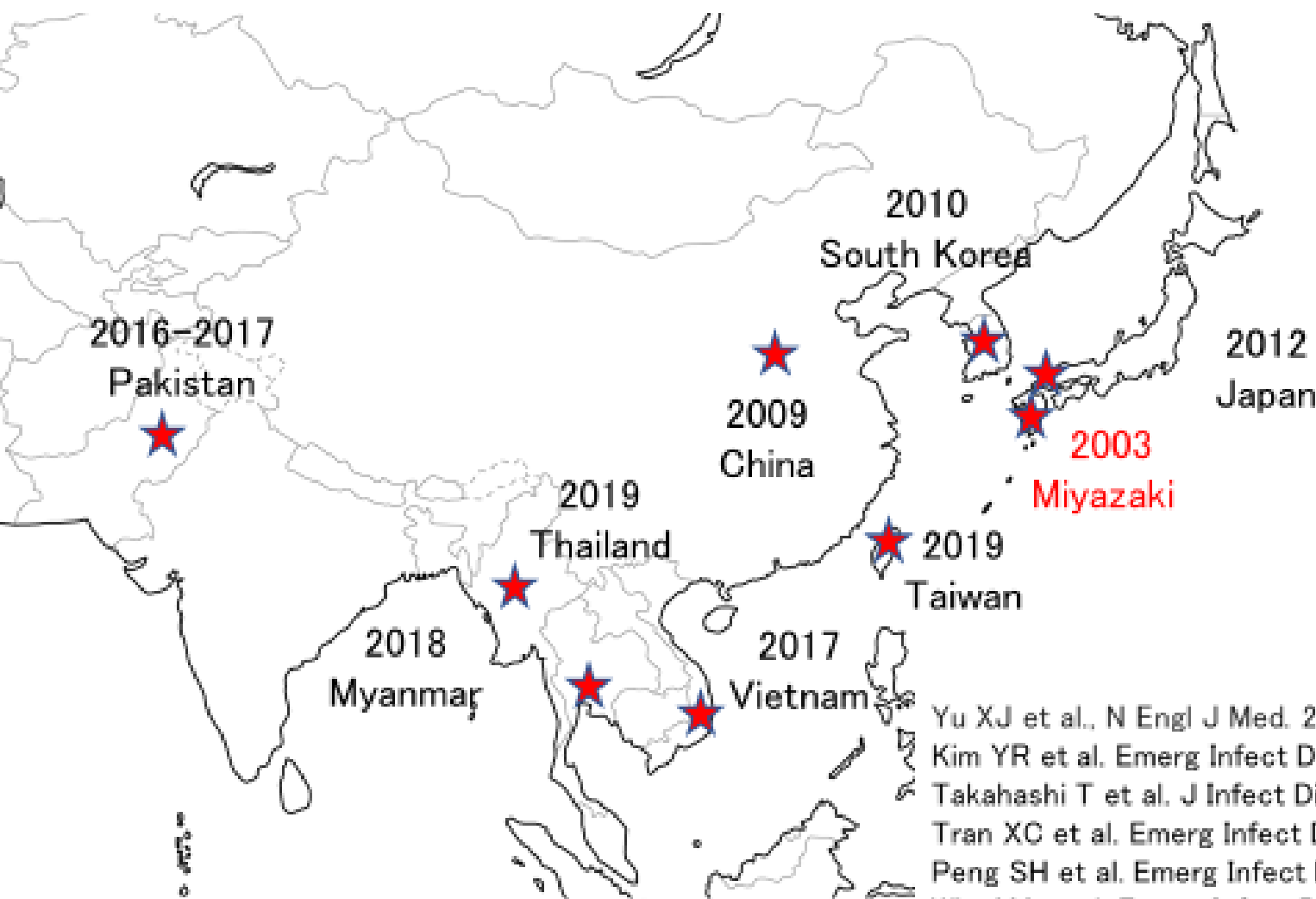
SFTSV transmission from a cat to humans



Direct transmission from animals to human

Jun, 2017	Dog⇒Owner
Aug, 2018	Cat⇒ Veterinarian and nurse
Oct, 2018	Cats⇒ Veterinarian
Oct, 2018	Cat⇒Owner (Dead)
May, 2019	Cat bite⇒Owner
Aug, 2019	Cat⇒Owner
Nov, 2019	Cat⇒ Veterinarian
Mar, 2020	Cat⇒Owner
Jun, 2020	Cat⇒ Veterinarian
Feb, 2021	Cat⇒ Veterinarian
Feb, 2021	Cat⇒ Veterinarian
Jun, 2021	Cat⇒ Veterinarian

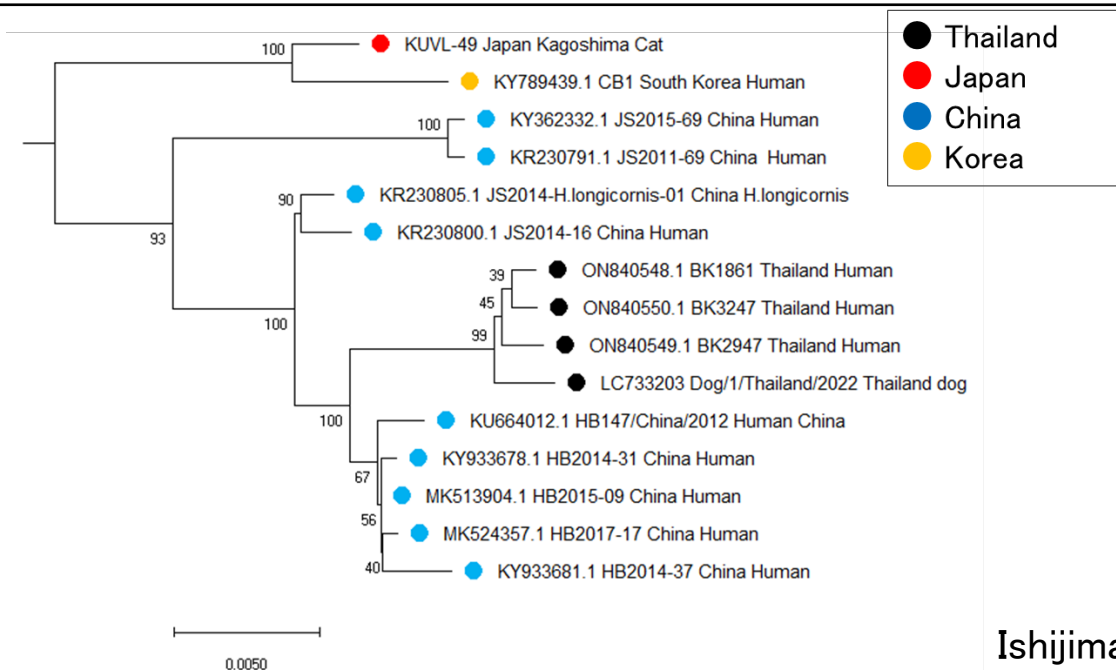
Distribution of SFTSV in Asian countries



- Yu XJ et al. *N Engl J Med*. 2011 Apr 21;364(16):1523-32
Kim YR et al. *Emerg Infect Dis*. 2018 Nov;24(11):2103-2105
Takahashi T et al. *J Infect Dis*. 2014 Mar;209(6):816-27
Tran XC et al. *Emerg Infect Dis*. 2019 May;25(5):1029-1031.
Peng SH et al. *Emerg Infect Dis*. 2020 Jul;26(7):1612-1614.
Win AM et al. *Emerg Infect Dis*. 2020 Aug;26(8):1878-1881
Ongkittikul S et al. *Bangkok Med J* 16, 2 (Sep. 2020), 204.
Zohaib A et al. *Emerg Infect Dis*. 2020 Jul;26(7):1513-1516.
Kirino Y et al. *J Infect Chemother*. 2022 Jun;28(6):753-756.

Prevalence of SFTSV infection in Thai dogs

Province	District	ELISA(OD>0.129)		FRNT ₅₀ (≥1:10)		Minimum Positive Ratio
		No. of Examined Dogs	No. of Positive Dogs	No. of Examined Dogs	No. of Positive Dogs	
Prachinburi		17	1	1	0	0.0%
Bangkok		143	12	12	6	4.2%
Chachoengsao		18	2	2	2	11.1%
Samutprakan		81	19	16	10	12.3%
Rayong		14	2	2	1	7.1%
Chonburi	Mueang Chonburi	27	6	6	3	11.1%
	Banglamung	56	5	5	3	5.4%
	Sattahip	95	56	52	50	52.6%
	Pattaya city	7	3	2	1	14.3%
Total		458	106	98	76	16.6%

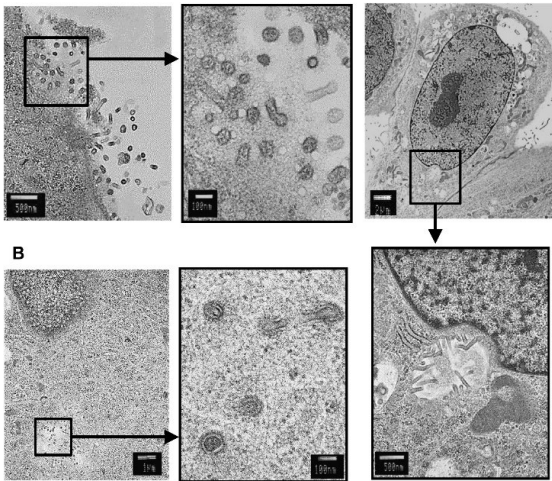


Summary of SFTS infection

- SFTS is a tick-borne disease
- SFTSV causes severe disease in human
- CFR in human is 27% in Japan
- SFTSV can infect to many animals
- SFTSV causes lethal disease in cats and the CFR is 60%
- SFTSV can transmit from diseased animals to humans
- Pet owners and veterinarians are at high risk of SFTSV infection
- Veterinarians should protect themselves by PPE (glove, mask, gown, goggles, face shield)

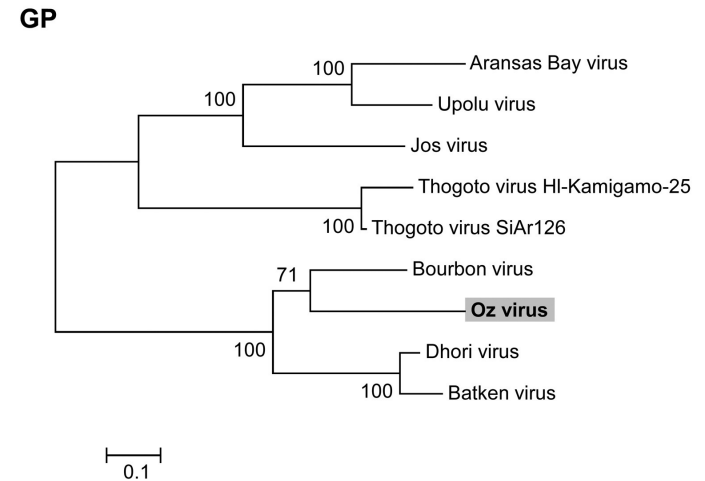
Characterization of Oz virus

A Oz virus belongs to *Paramyxoviridae*

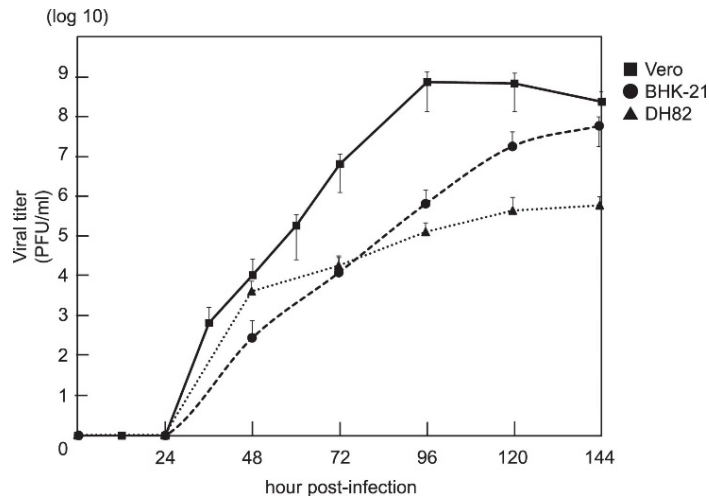


Pleomorphic viral particles, including both filamentous and round particles, budded directly from the plasma membrane of infected cells

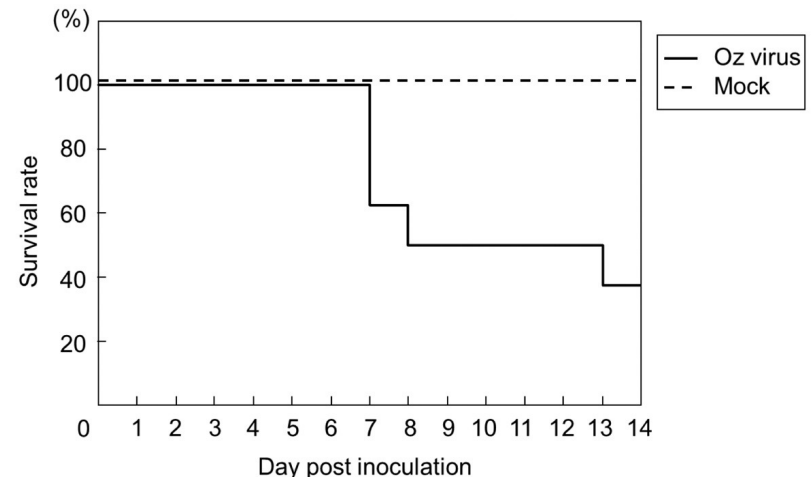
Oz virus is similar to Bourbon virus in U.S.A



Oz virus can grow well in mammalian cell lines



Oz virus can kill suckling mice



Oz virus infects many mammalian species

Species	Year	VN titer						Ratio	
		<1:10	1:10	1:20	1:40	1:80	1:160		1:320
Human	2015	22	0	0	1	1	0	0	8.3%
Monkey	2012– 2019	91	0	4	5	4	9	7	24.2%
Wild boar	2013– 2015	49	2	12	10	15	20	16	60.5%
Sika deer	2014– 2015	20	5	8	11	12	13	7	73.7%

The first patient with Oz virus infection

Date: early summer of 2022

Patient: female in her **70s**, resident of Ibaraki Prefecture, underlying hypertension and dyslipidemia, **no history of overseas travel**

Symptoms: **fatigue, loss of appetite, vomiting, joint pain, and a fever of 39 °C**, ultimately resulting in **viral myocarditis and death**

Blood tests: **thrombocytopenia** (66,000/ μ L), **liver disorder**, renal disorder, high serum markers of inflammation (CRP 22.82 mg/dL), **high CK** (2049 U/L, CK-MB 14 IU/L), **high LDH** (671 U/L), **high ferritin** (10729 ng/mL), simple CT normal.

On hospitalization: **Engorged ticks** in the right inguinal region. No rickettsia or SFTS, blood culture negative.

During hospitalization: Pacemaker implanted due to atrioventricular block. Various tests suspected myocarditis. The pacemaker was removed after approximately 10 days, when the patient's pulse stabilized. On the 20th day of hospitalization, consciousness disorder and multiple cerebral infarctions were observed, and anticoagulation therapy was started. The patient had persistent fever, but thoracoabdominal pelvic contrast CT could not point out any obvious foci or organ enlargement that could be the source of the fever.

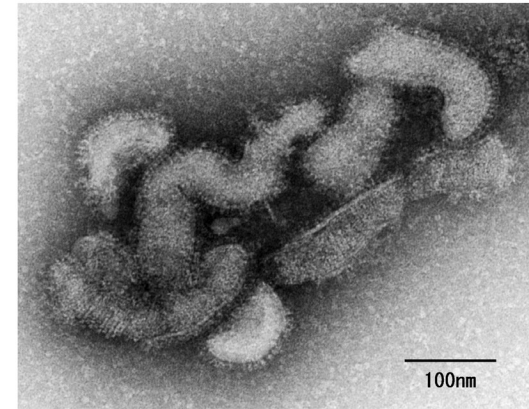


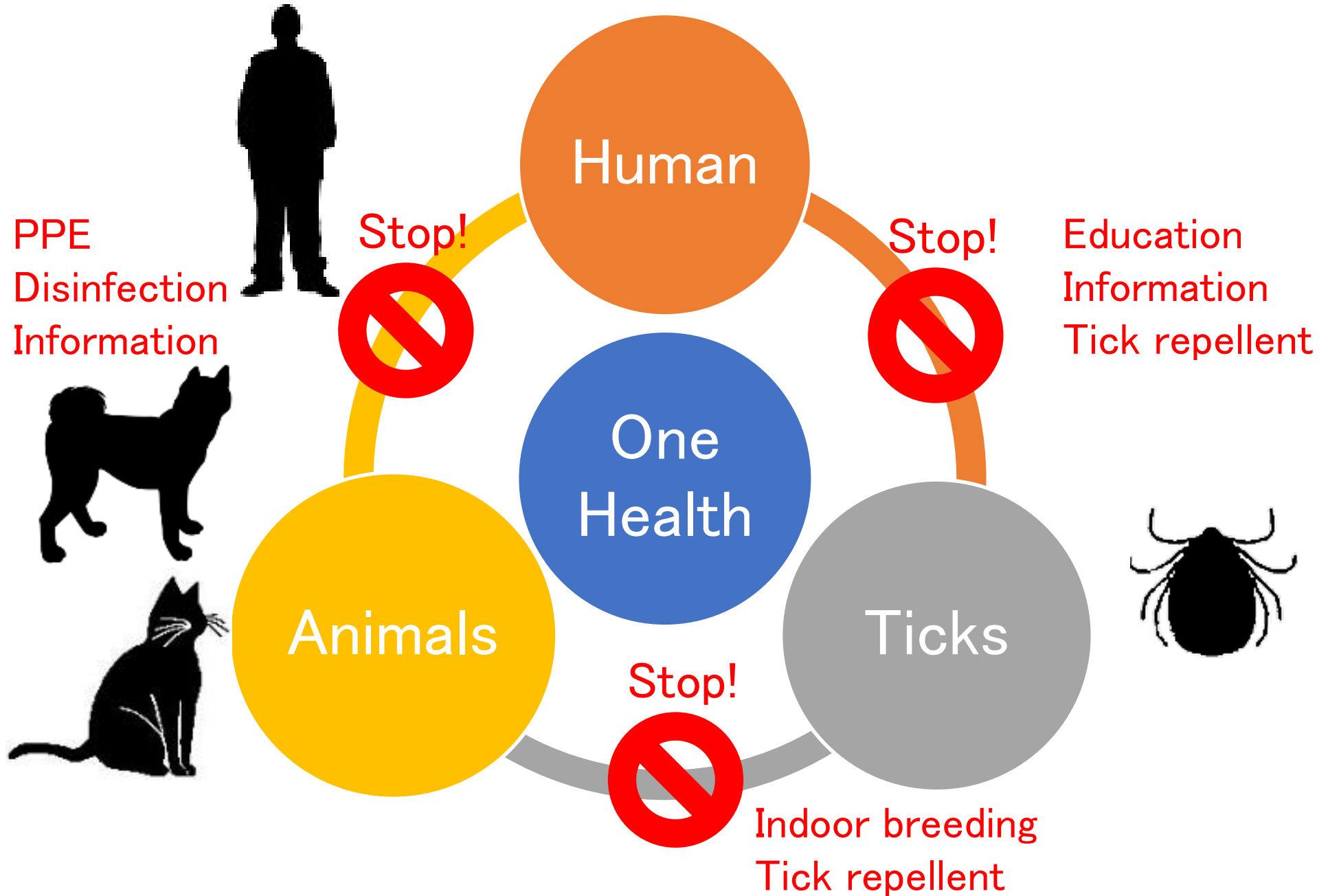
図. 患者検体から分離されたOZV粒子の電子顕微鏡写真

IASR

Catalogue of tick-borne viruses in Japan

Viruses	Classification	Isolation	Tick	Animal	Human	Disease
SFTS virus	<i>Bunyavirales</i>	○	○	○	○	SFTS
Yezo virus	<i>Bunyavirales</i>	○	○	○	○	Like SFTS
Iwanai Valley virus	<i>Bunyavirales</i>	○	○			
Ama virus	<i>Bunyavirales</i>	○	○			
Tofla virus	<i>Bunyavirales</i>	○	○			
Toyo virus	<i>Bunyavirales</i>		○			
Kabuto Mountain virus	<i>Bunyavirales</i>	○	○	○	○	?
Okutama tick virus	<i>Bunyavirales</i>		○			
Mukawa virus	<i>Bunyavirales</i>	○	○	○		
Soft tick bunyavirus	<i>Bunyavirales</i>	○	○			
Kuriyama virus	<i>Bunyavirales</i>	○	○			
Tick-borne encephalitis virus	<i>Flaviviridae</i>	○	○	○	○	Encephalitis
Yamaguchi virus	<i>Flaviviridae</i>		○	○		
Negishi virus	<i>Flaviviridae</i>	○			○	Encephalitis
Saruyama virus	<i>Flaviviridae</i>		○	○		
Ohshima virus	<i>Orthomyxoviridae</i>		○			
Thogoto virus	<i>Orthomyxoviridae</i>	○	○	○	?	
Oz virus	<i>Orthomyxoviridae</i>	○	○	○	○	Myocarditis
Tarumizu tick virus	<i>Reoviridae</i>	○	○	○		
Muko virus	<i>Reoviridae</i>	○	○	○		
Kemerovo virus	<i>Reoviridae</i>	○	○	?	?	
Sekira virus	<i>Nyamiviridae</i>		○			
H. Flava Ifla virus	<i>Iflaviridae</i>		○			

One Health approach for tick-borne diseases



Acknowledgements



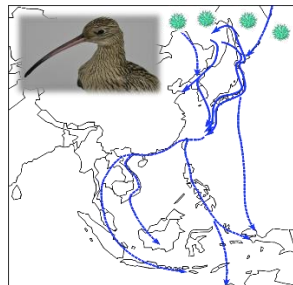
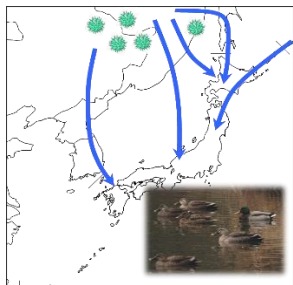
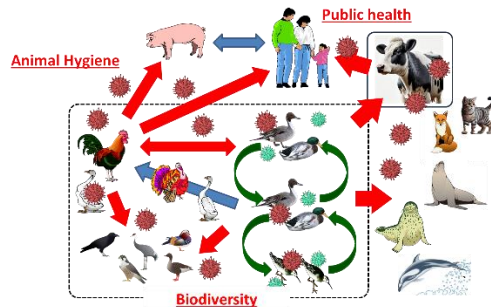
Yamaguchi University
Kagoshima University
Nagasaki University
Okayama Science University
Miyazaki University
Tokyo University of Agriculture and
Technology
The University of Tokyo
Hokkaido University
Hiroshima Veterinary Medical Association

Veterinary Professionals throughout Japan

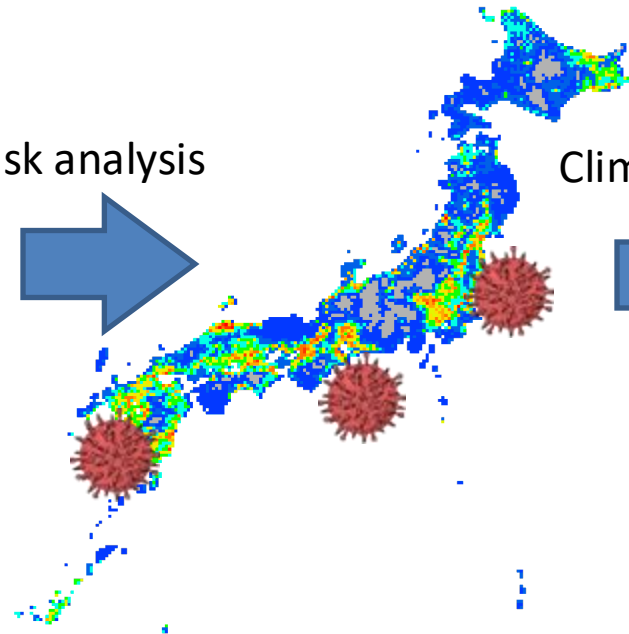
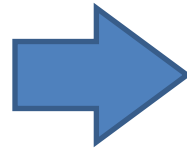
National Institute of Infectious Diseases
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Department of Pathology
Department of Medical Entomology
Research Center for Influenza and
Respiratory Viruses
Research Center for Biosafety,
Laboratory Animal and Pathogen Bank



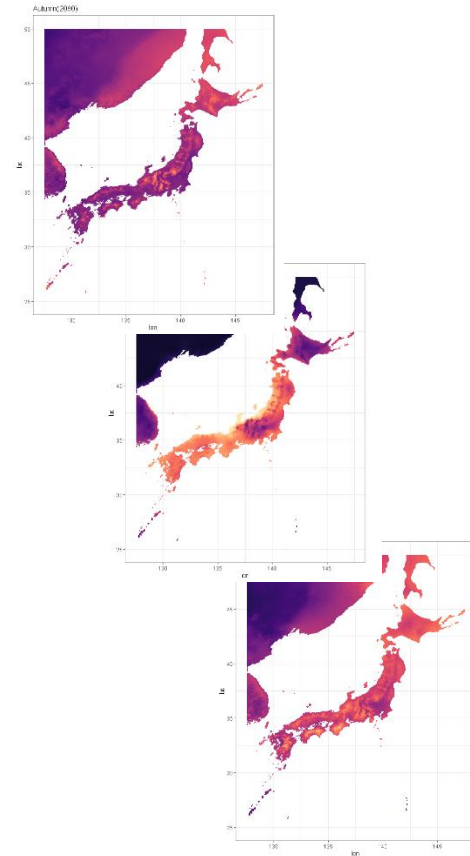
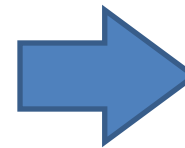
Climate change, wildlife and zoonoses: HPAIV as a model case



Risk analysis



Climate change



Koichi GOKA and Manabu Onuma
National Institute for Environmental Studies

Wild Ducks as Long-Distance Vectors of Highly Pathogenic Avian Influenza Virus (H5N1)

Juthatip Keawcharoen,* Debby van Riel,* Geert van Amerongen,* Theo Bestebroer,* Walter E. Beyer,* Rob van Lavieren,* Albert D.M.E. Osterhaus,* Ron A.M. Fouchier,* and Thijs Kuiken*

Emerging Infectious Diseases • www.cdc.gov/eid • Vol. 14, No. 4, April 2008

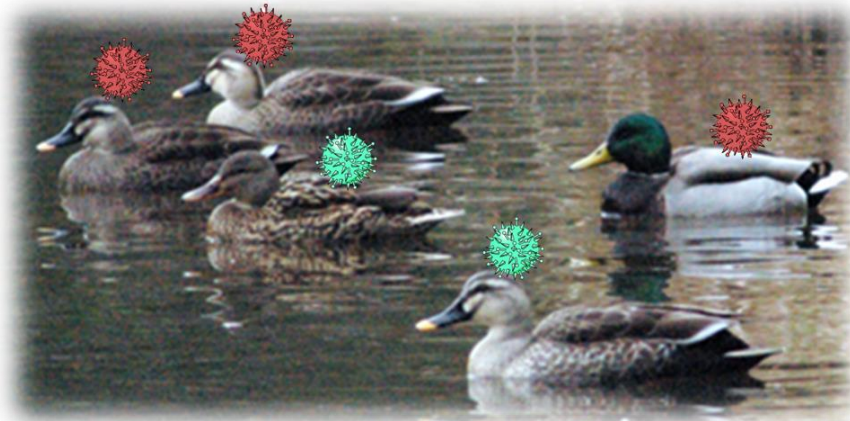
Comment

<https://doi.org/10.1038/s41564-023-01538-0>

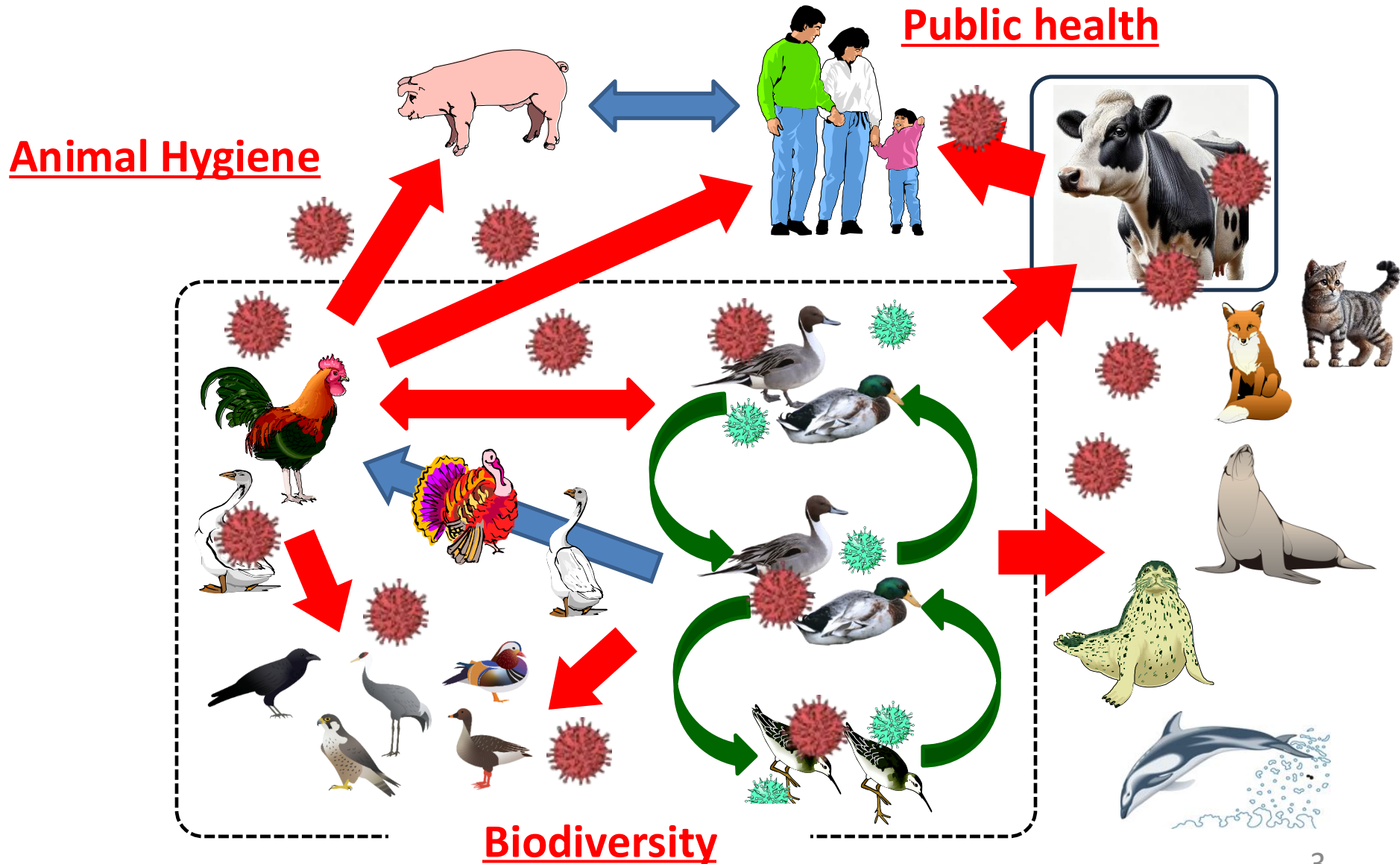
Climate change impacts on bird migration and highly pathogenic avian influenza

Diann J. Prosser, Claire S. Teitelbaum, Shenglai Yin, Nichola J. Hill & Xiangming Xiao

Check for updates

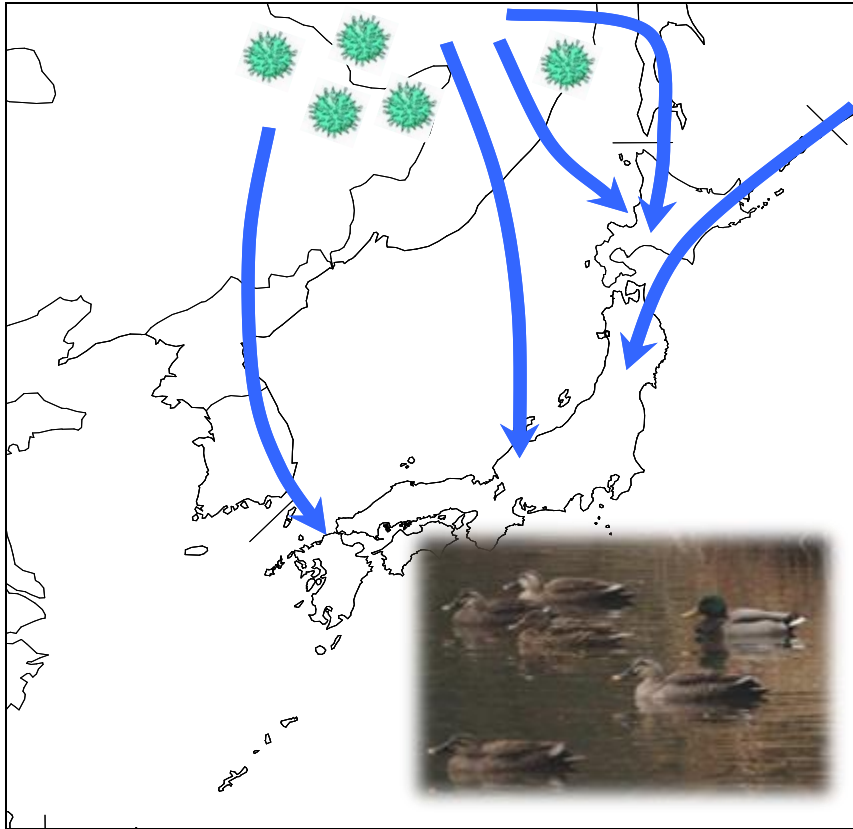


Animals infected with highly pathogenic avian influenza viruses



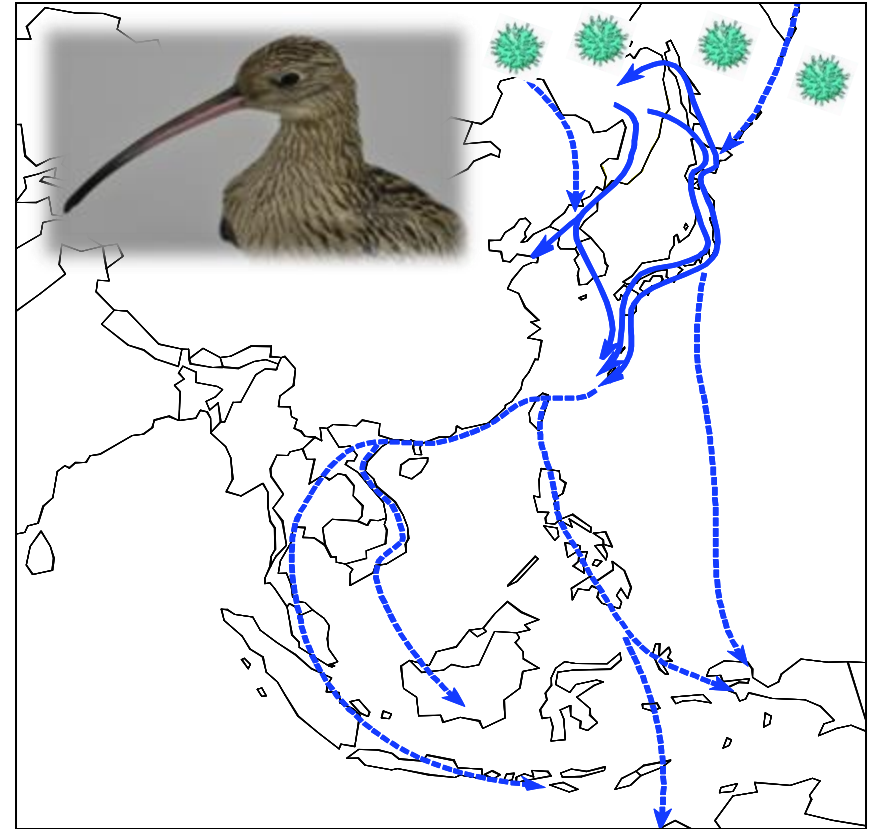
Nationwide AIV surveillance in Japan

Wild ducks
(wintering in Japan)
(October – March)



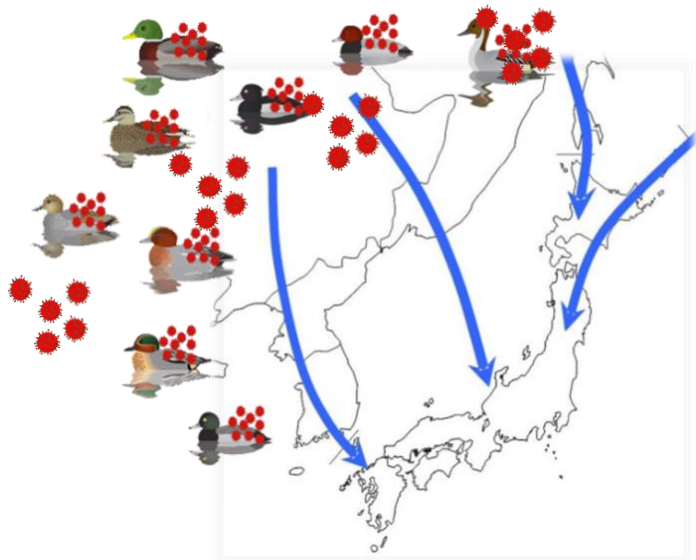
Funded by MOE

Shorebird
(The East-Asian Australasian Flyway)
(July – September)



Funded by NIES

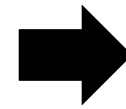
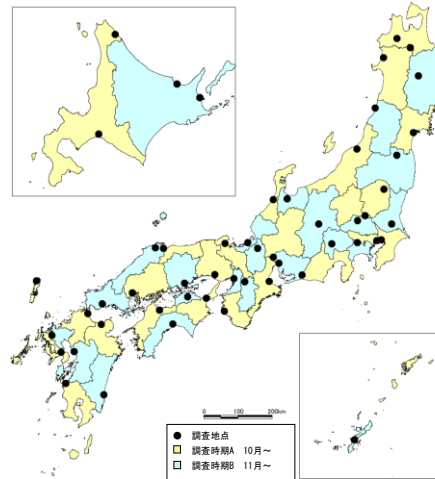
Nationwide AIV surveillance using fecal samples of Wild ducks (2008-2020)



52 locations
(Oct.-May)

≈3,000 tubes
/season

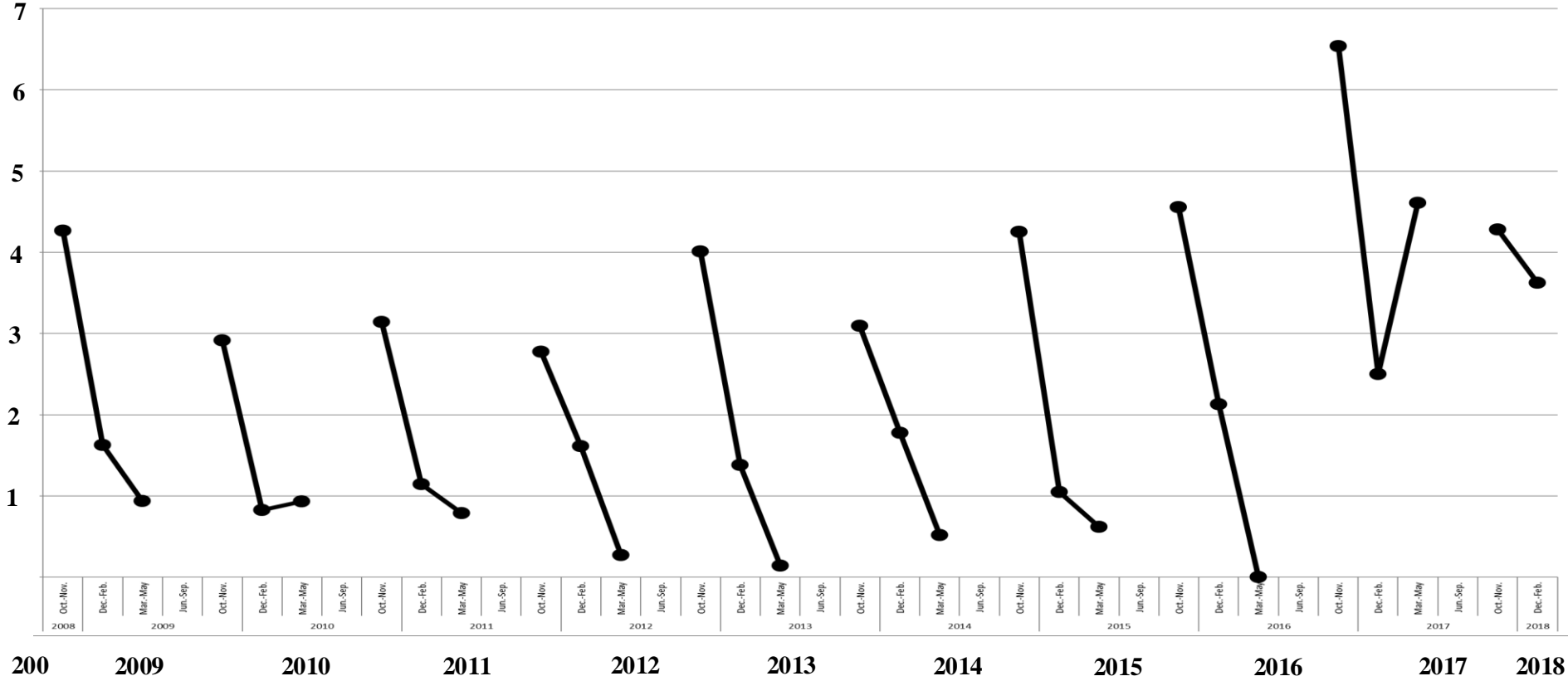
5 feces/tube



Temporal change of AIV prevalence (2008-2018)

RT-LAMP positive rate in fecal samples (%)

Onuma M. Unpublished data



2008 2009 2010 2011 2012 2013 2014 2015 2016 2017 2018

↔ H5N1 ↔
↔ H5N8 ↔
↔ H5N6 ↔
↔ H5N6 ↔

HPAI cases of wild birds in Japan (2004-2011)
H5N1: 17 species & 1 unknown species, 75 individuals



HPAI cases of wild birds in Japan (2014-2015)
H5N8: 4 species & 1 unknown species, 9 individuals



HPAI cases of wild & captive birds in Japan (Oct.2016-Apr.2017)
H5N6: 25 species & 1 unknown species, 210 individuals



HPAI cases of wild & captive birds in Japan (Nov.2017-Mar.2018)
H5N6: 5 species, 46 individuals



Time series changes in positivity rates (by region)



Autum migration
(Oct. ~ Nov.)



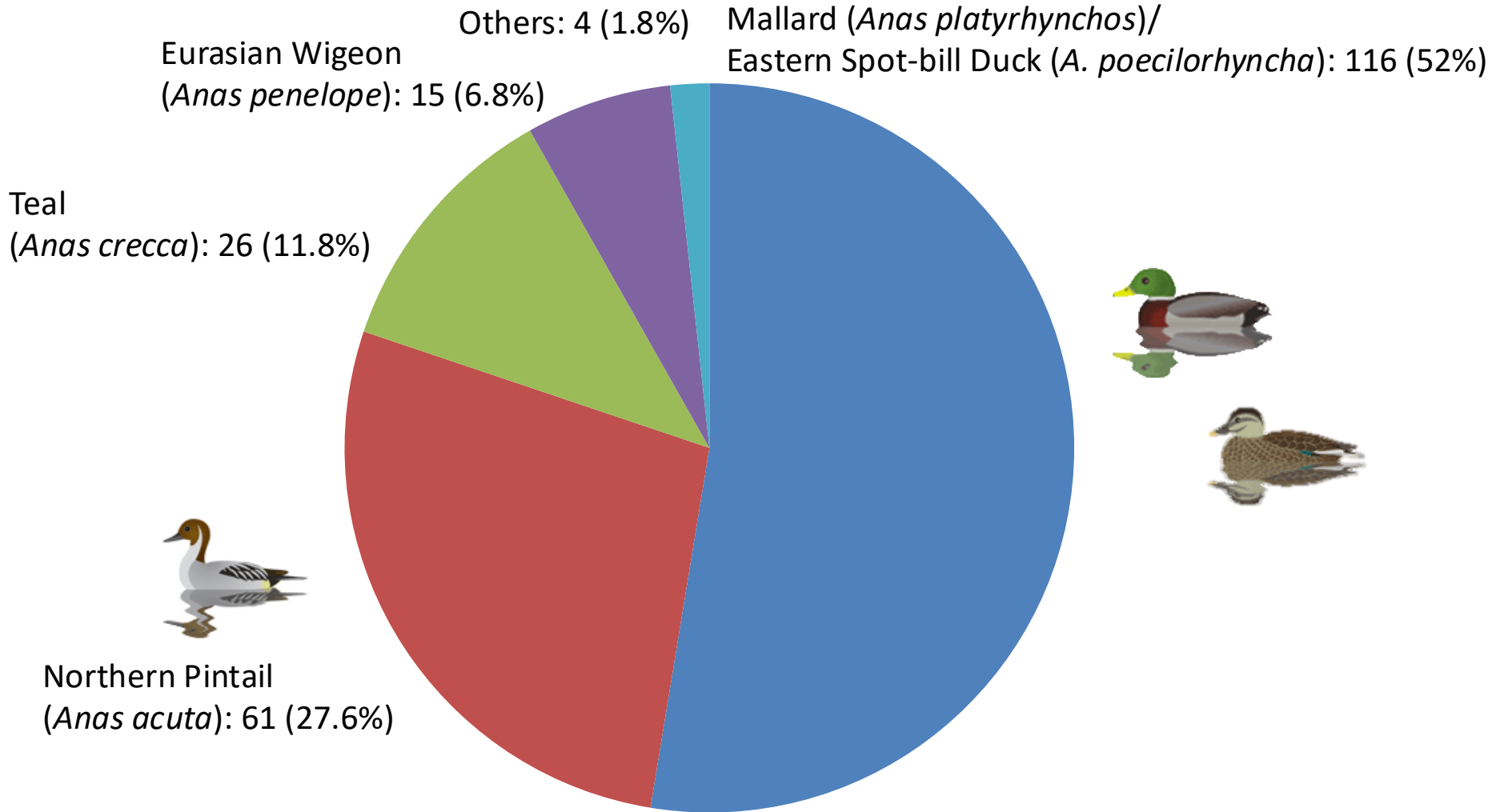
Wintering
(Dec. ~ Feb.)



Spring migration
(Mar. ~ May)

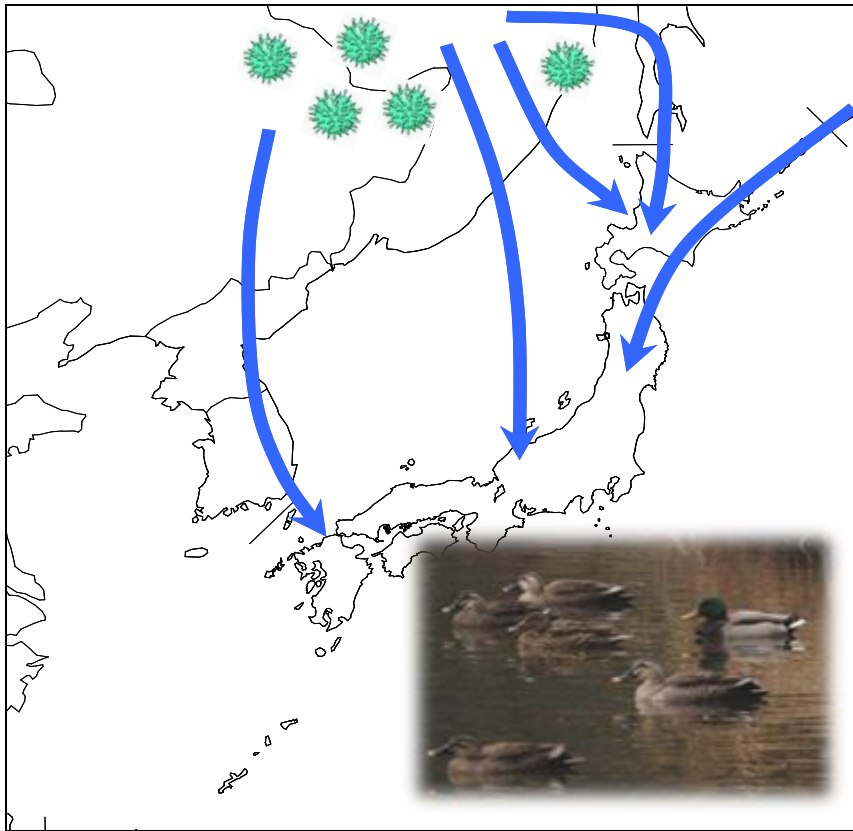
 Regions with positivity rates above the national average

Result of species identification by DNA barcoding



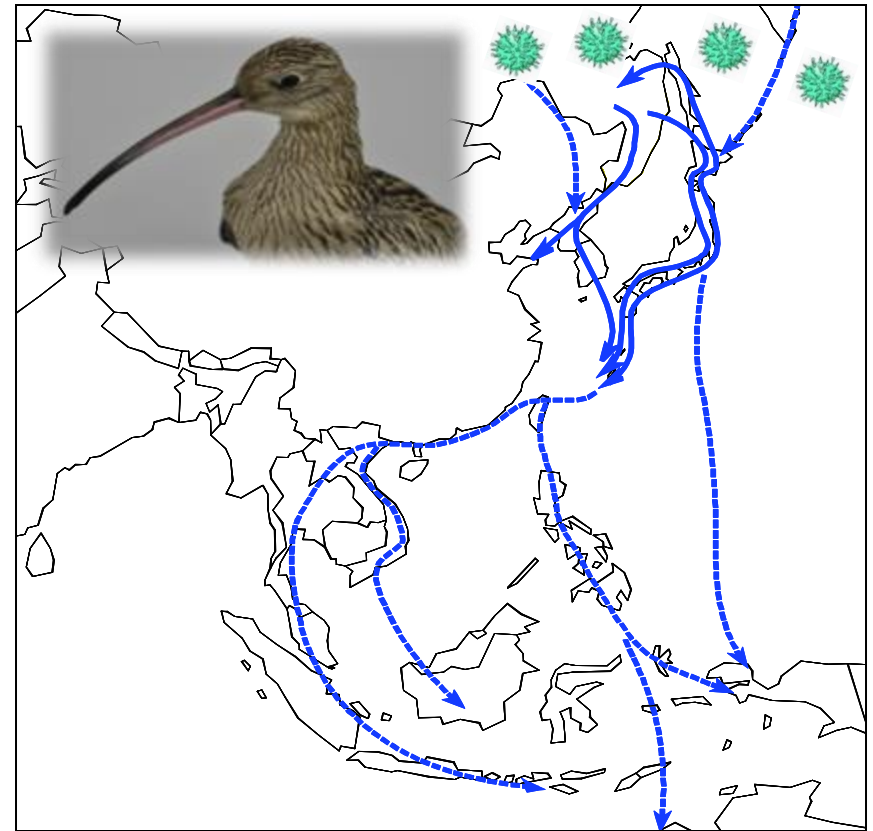
Nationwide AIV surveillance in Japan

Wild ducks
(wintering in Japan)
(October – March)



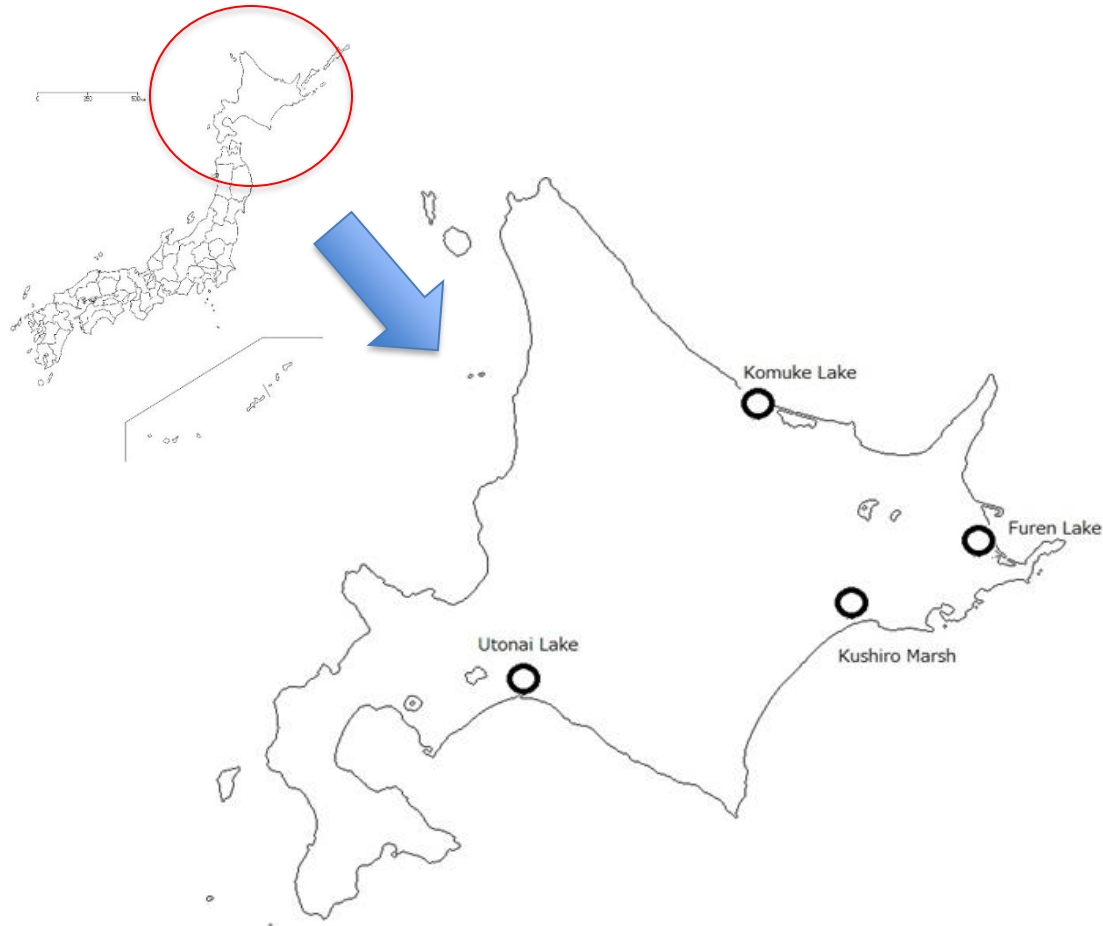
Funded by MOE

Shorebird
(The East-Asian Australasian Flyway)
(July – September)



Funded by NIES

Nationwide AIV surveillance in shorebirds



Result: AIV detection (by RT-LAMP)

	2006	*2007	*2008	2009	*2010	Total
Number	340	289	210	329	579	1,747
Positive (%)	0	0	0	0	1 (0.2)	1 (0.1)

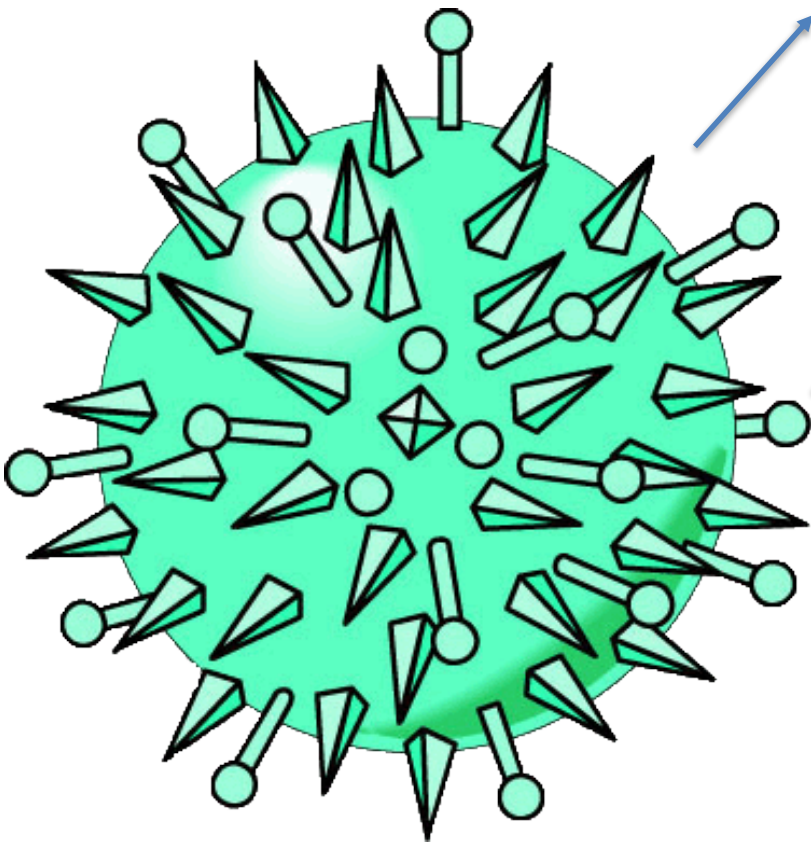
* HPAIV positive cases in wild birds



Lesser Sand Plover
(*Charadrius mongolus*)

Captured at **Komuke lake**
in **September 3rd**, 2010

Result: Subtyping (by sequencing)



HA: **H10** (1,686 bp)
A/mallard/Korea/1242/2010(H10N6)
(99% identity)

NA: **N7** (1,416 bp)
A/common teal/Hong Kong/
MPM1740/2011(H7N7) (99% identity)

Antibody prevalence of avian influenza virus in shorebirds (2017~)



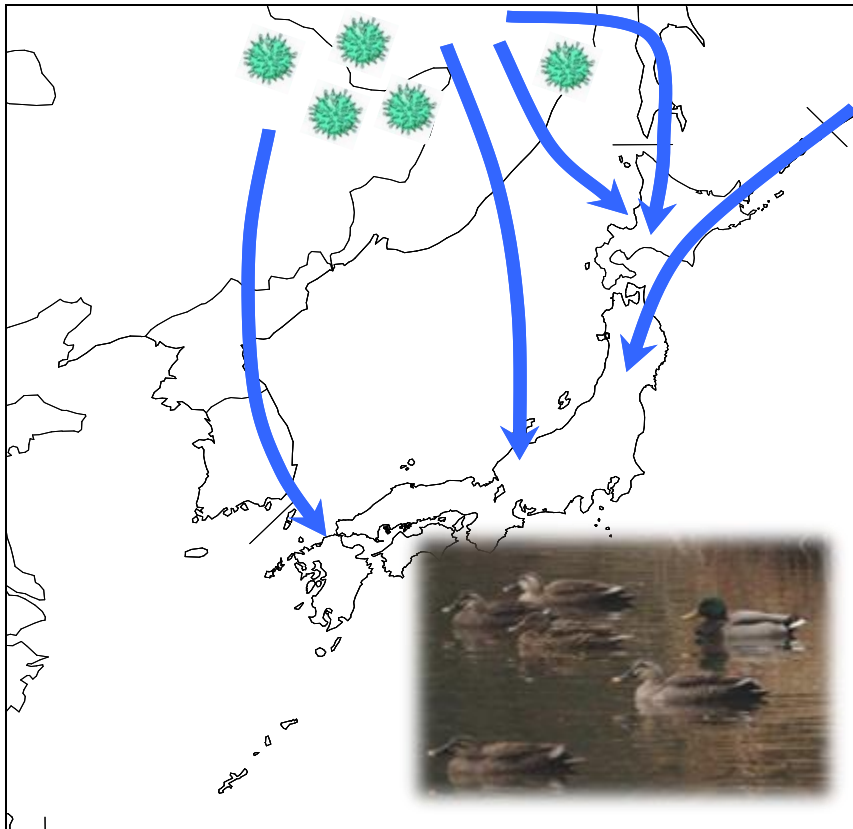
Ruddy Turnstone
(Arenaria interpres)
34.8% (8/23)



Whimbrel
(Numenius phaeopus)
36.4% (4/11)

Nationwide AIV surveillance in Japan

Wild ducks
(wintering in Japan)
(October – March)



Funded by MOE

Shorebird
(The East-Asian Australasian Flyway)
(July – September)



Funded by NIES

Identification of high-risk regions



Autum migration
(Oct. ~ Nov.)

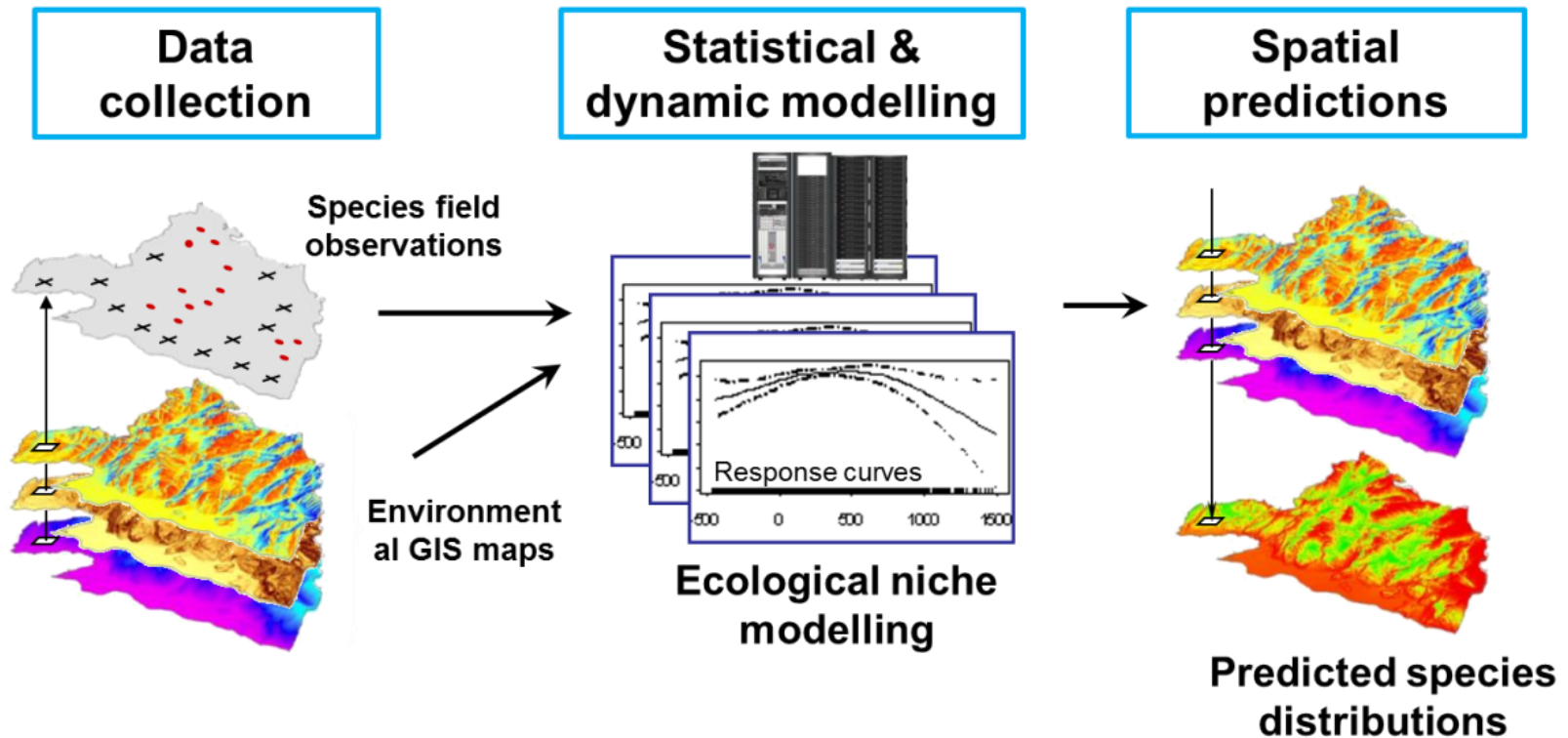


Wintering
(Dec. ~ Feb.)



Spring migration
(Mar. ~ May)

Environmental niche modelling

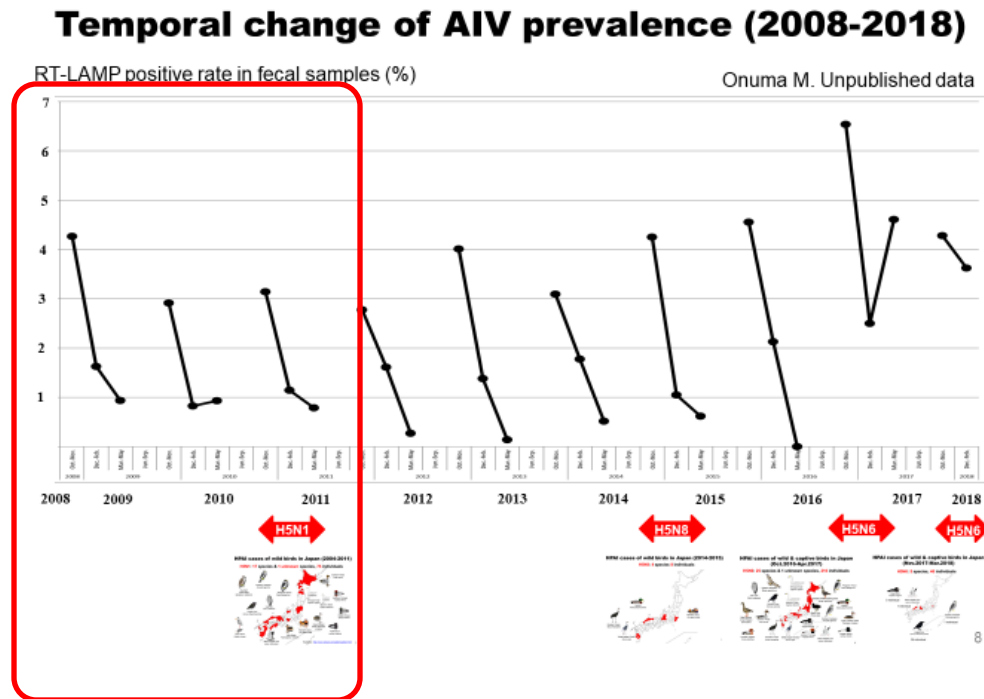


<https://www.unil.ch/webdav/site/idyst/shared/modeling.png>

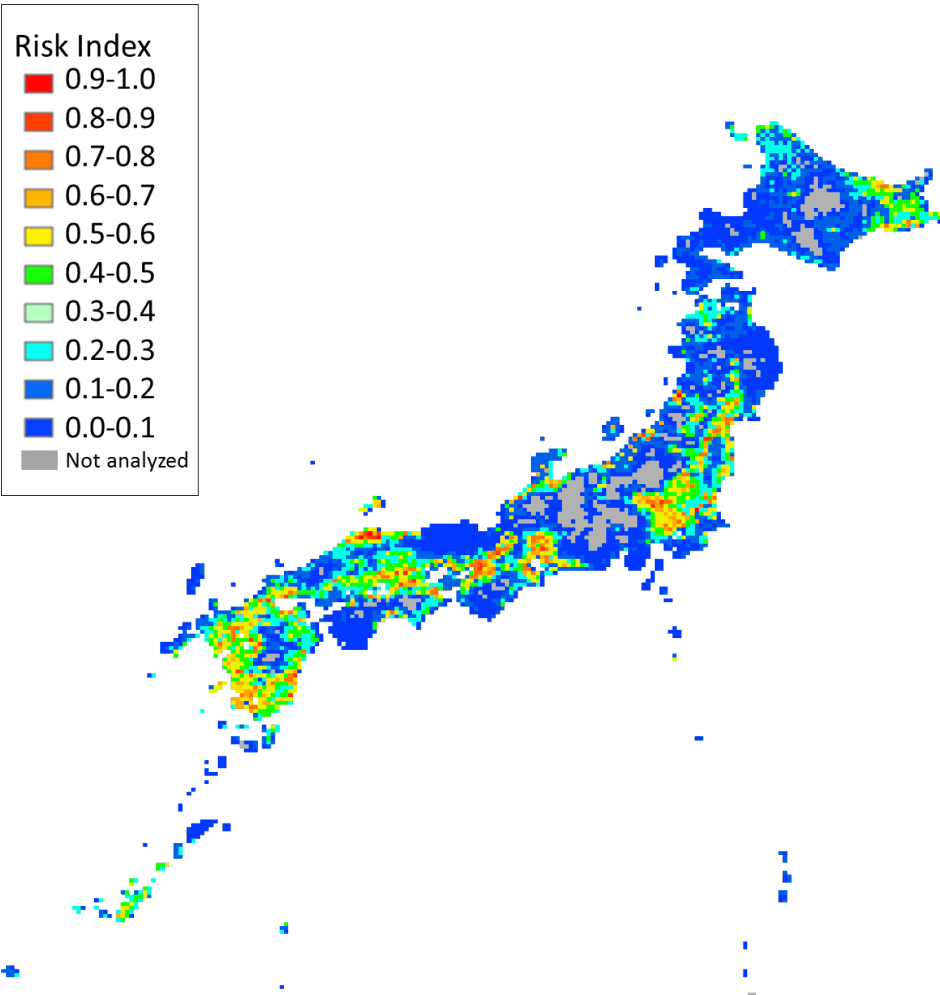
Method

Program: MaxEnt version 3.3.3e, Phillips et al., 2006
(The maximum entropy approach)

Data: Locations of LPAIV and HPAIV 2004-2011



Risk map for avian influenza in wild birds



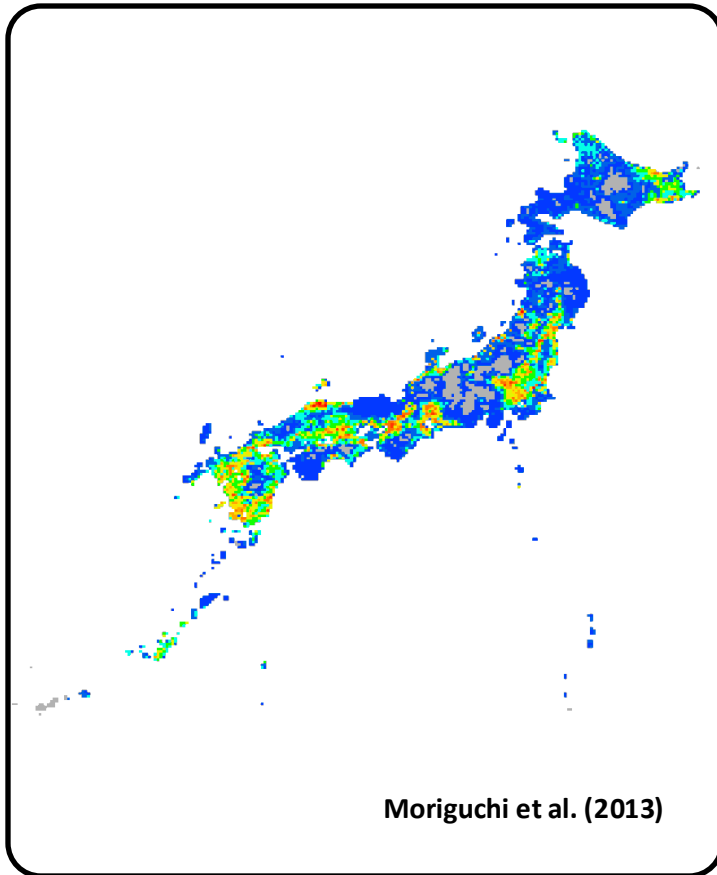
Contributions of independent variables to the model.

Independent variables	Contribution
Dabbling duck population	49.4 ± 5.5
Urban area	12.5 ± 3.5
Altitude	8.2 ± 2.3
Diving duck population	3.5 ± 3.5
Lake area	2.2 ± 1.5
Farmland area	0.9 ± 1.2
Poultry density	0.5 ± 0.5
Mean precipitation in winter	0.3 ± 1.0
Min temperature in winter	0.1 ± 0.1
Distance to lakes	0.0 ± 0.0
Sum of spatial filters	22.5 ± 5.6

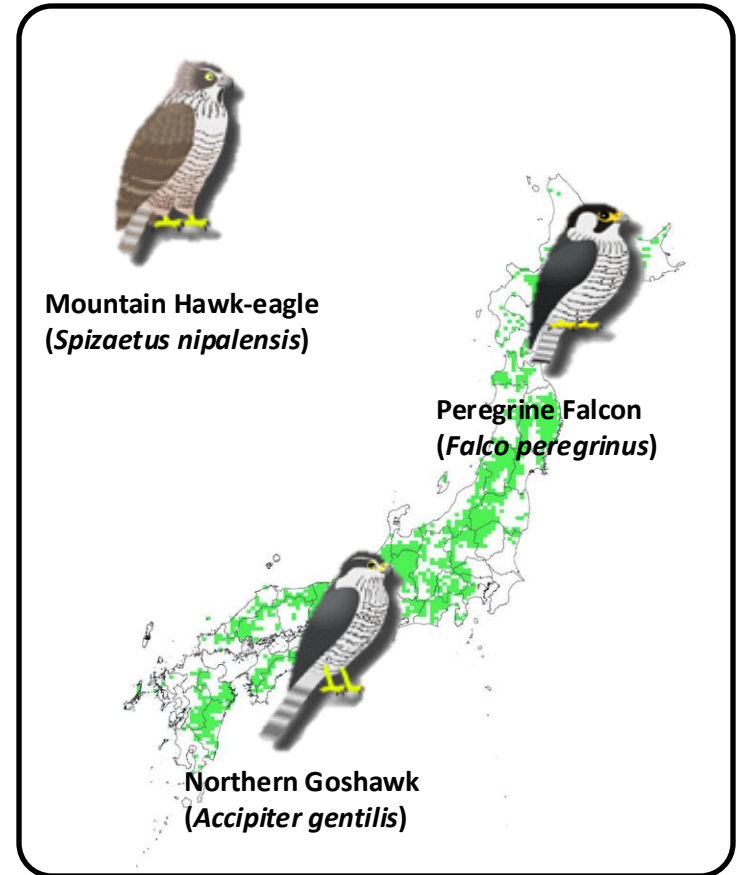
(Moriguchi et. al., 2013)

Risk map of avian influenza in raptors

Risk map of AI



Distribution map



Risk map of avian influenza in raptors



Mountain Hawk-eagle
(*Spizaetus nipalensis*)

HPAIV:1



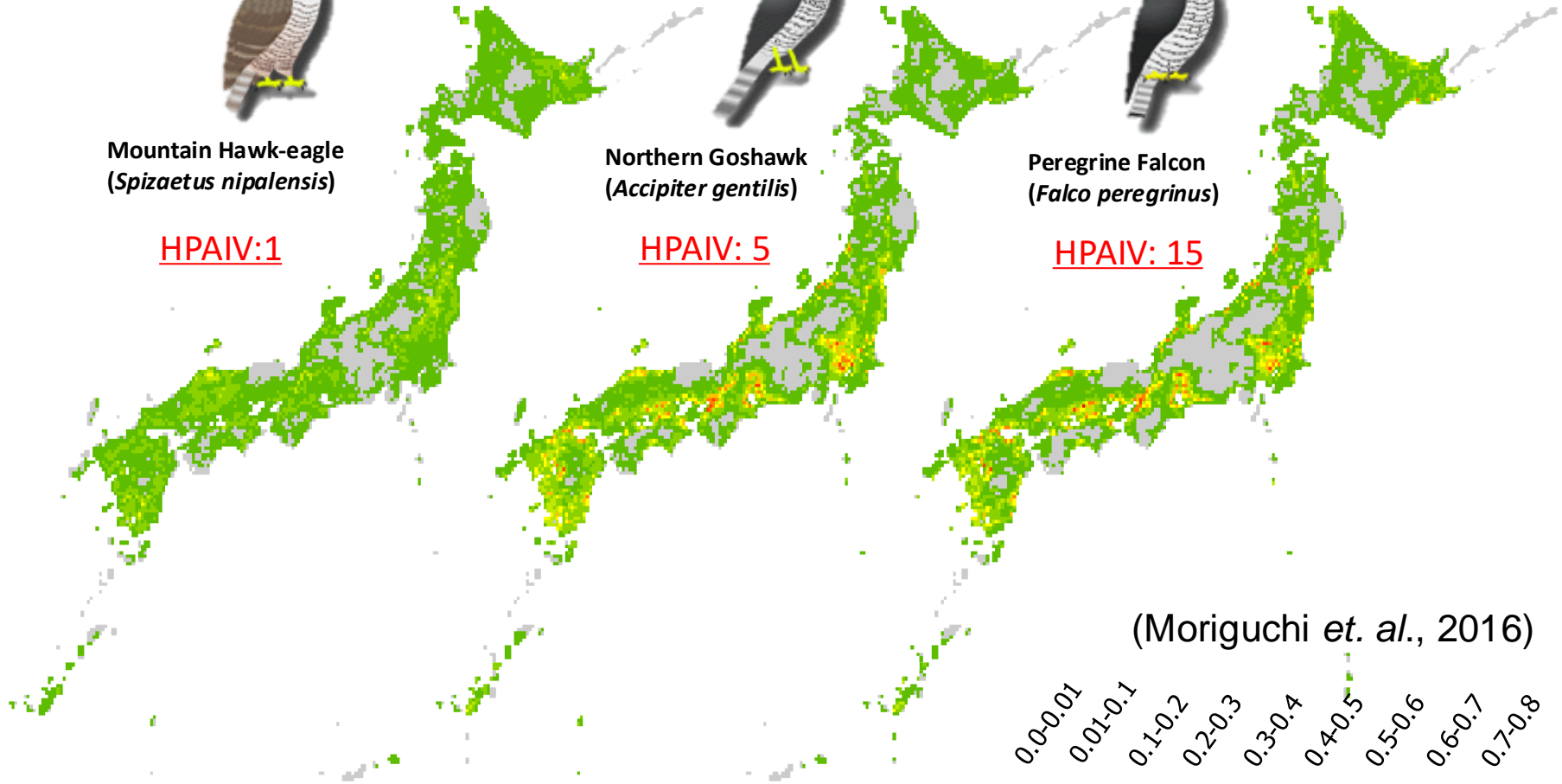
Northern Goshawk
(*Accipiter gentilis*)

HPAIV: 5

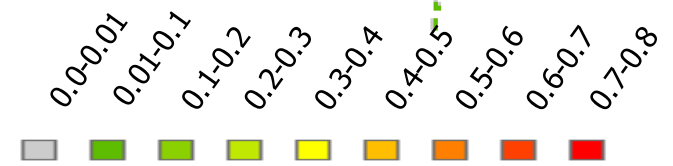


Peregrine Falcon
(*Falco peregrinus*)

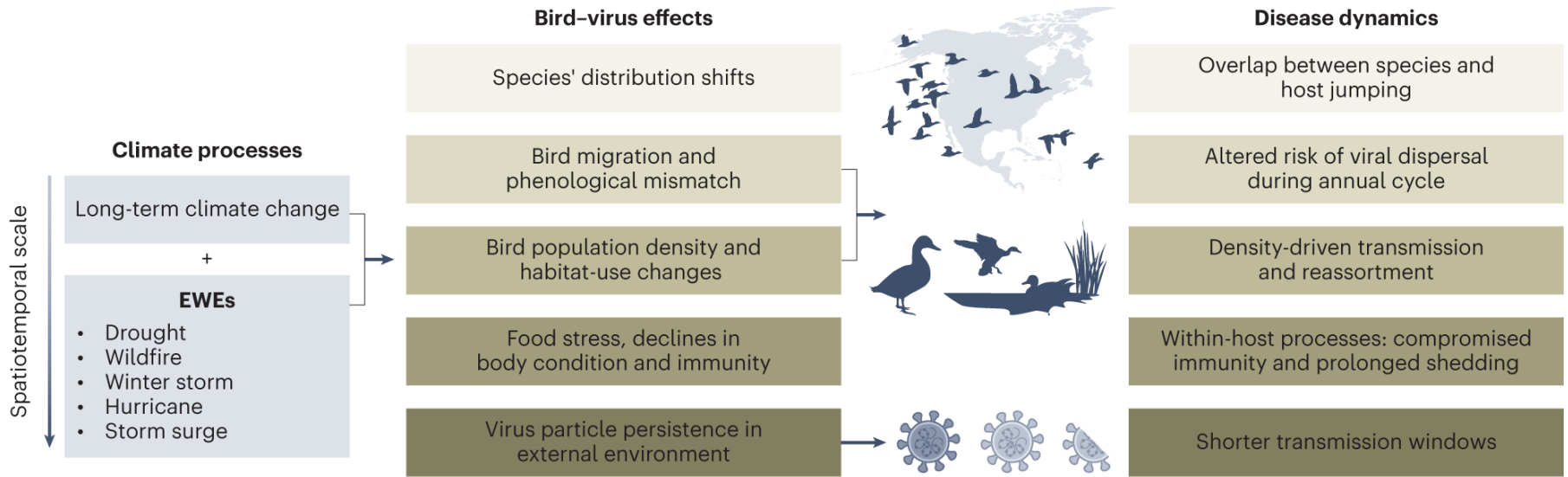
HPAIV: 15



(Moriguchi *et. al.*, 2016)



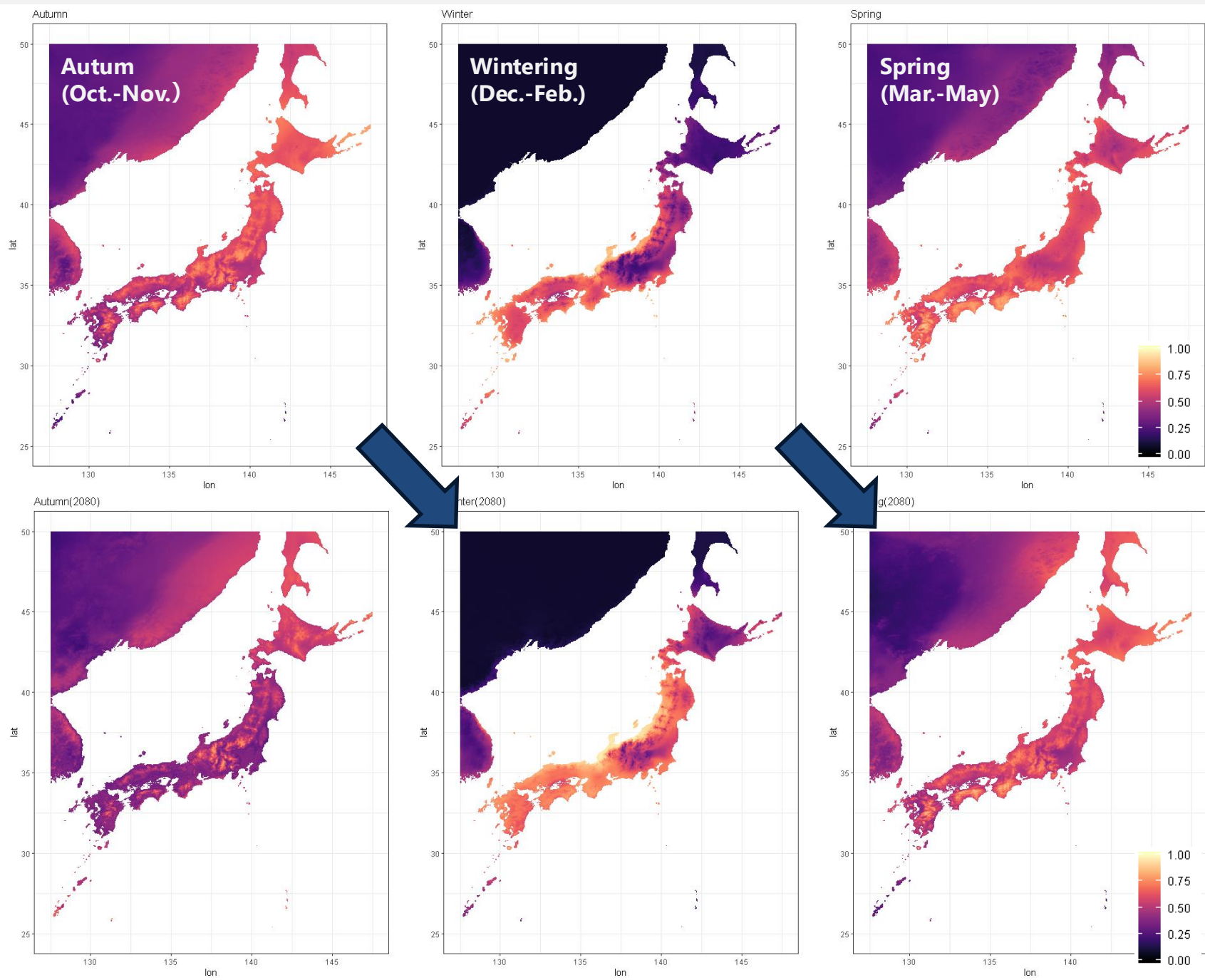
Climate change impacts on bird migration and highly pathogenic avian influenza (Prosser et al. 2023)



doi: 10.1038/s41564-023-01538-0.

" long-distance migratory species tend **to track their niche** and select largely **similar environments through seasons**" Zurell et al. (2018, J. Biogeog.)

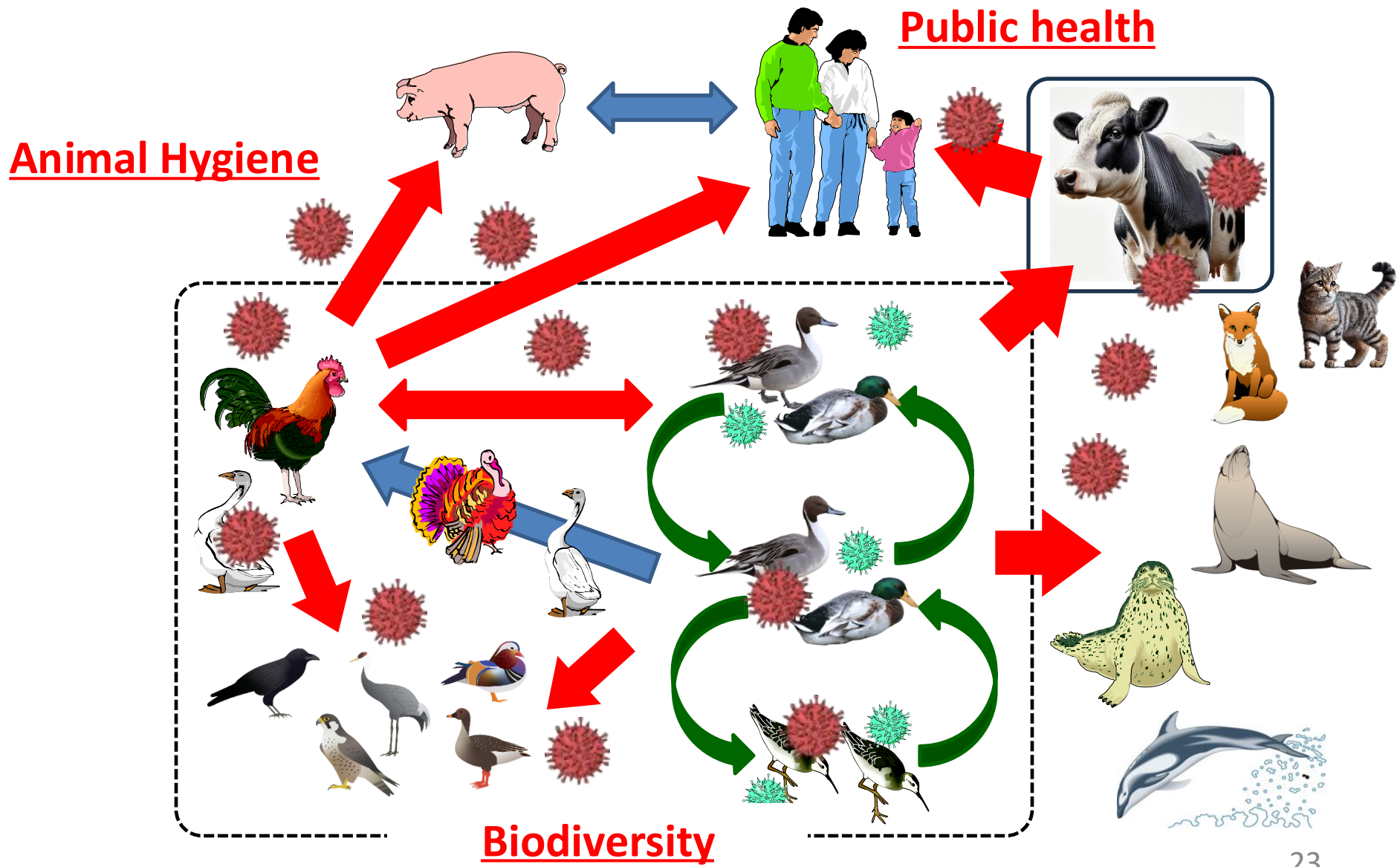
"Is it possible to estimate 'when' and 'where' migratory birds are using an ecological niche model with seasonal climate data?"



Present

**2080
(RCP8.5)**

Animals infected with highly pathogenic avian influenza viruses



Mass mortality event of marine mammals in Russia (Summer 2023)

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