



**Albert Osterhaus DVM PhD**

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# **VIRUSES of wildlife: Elephants and Rhinos**



**Blijdorp Zoo  
Rotterdam  
October 11th 2011**





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**DECLARATION OF INTEREST/ EMPLOYMENT:**

**Professor/Head Department of Virology Erasmus University/MC (NL)**

**Professor Virology Utrecht University (NL)**

**Chair Artemis Wildlife Health Institute (Europe)**

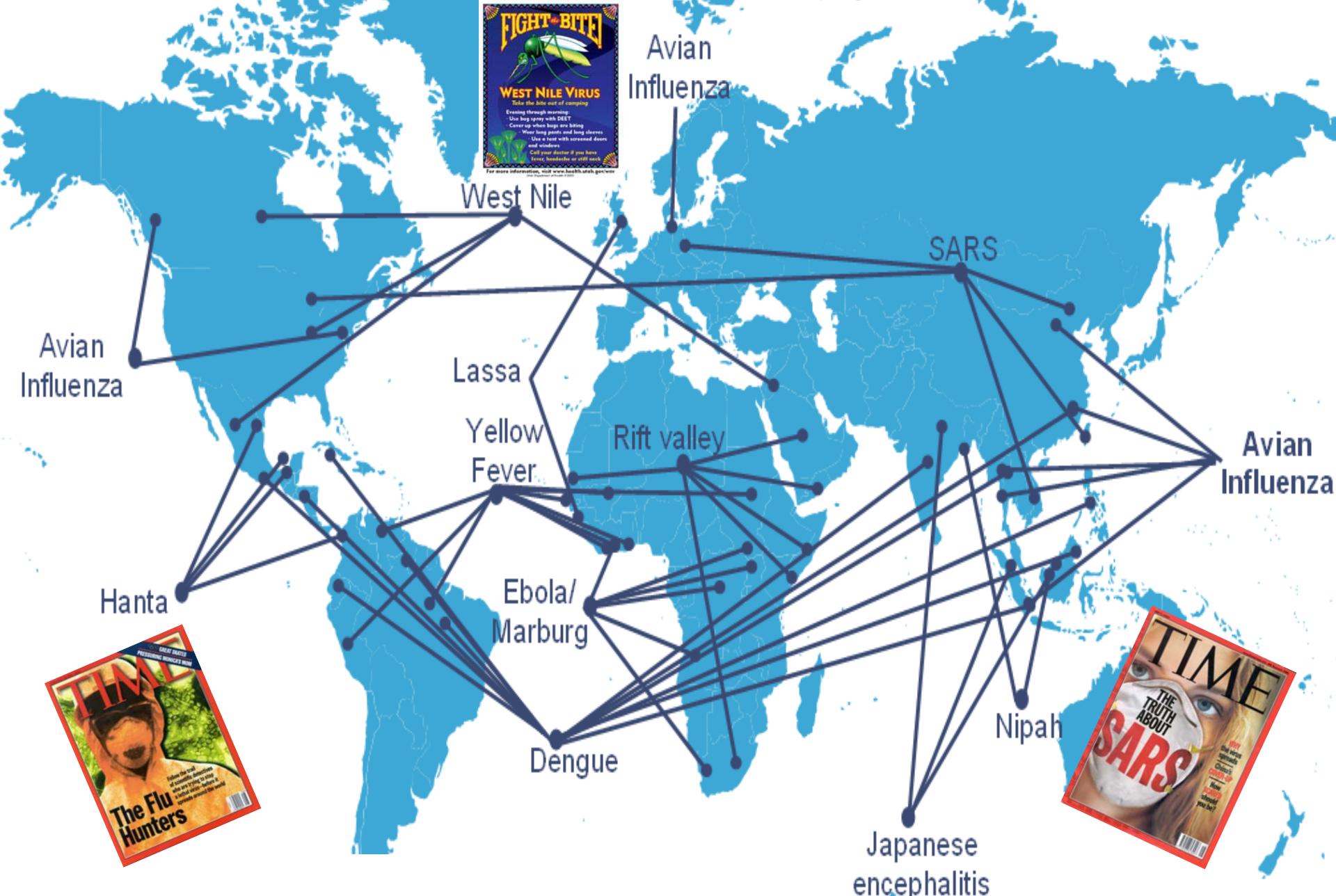
**CSO Viroclinics-Biosciences BV (NL)**

**Chair ESWI (Europe)**



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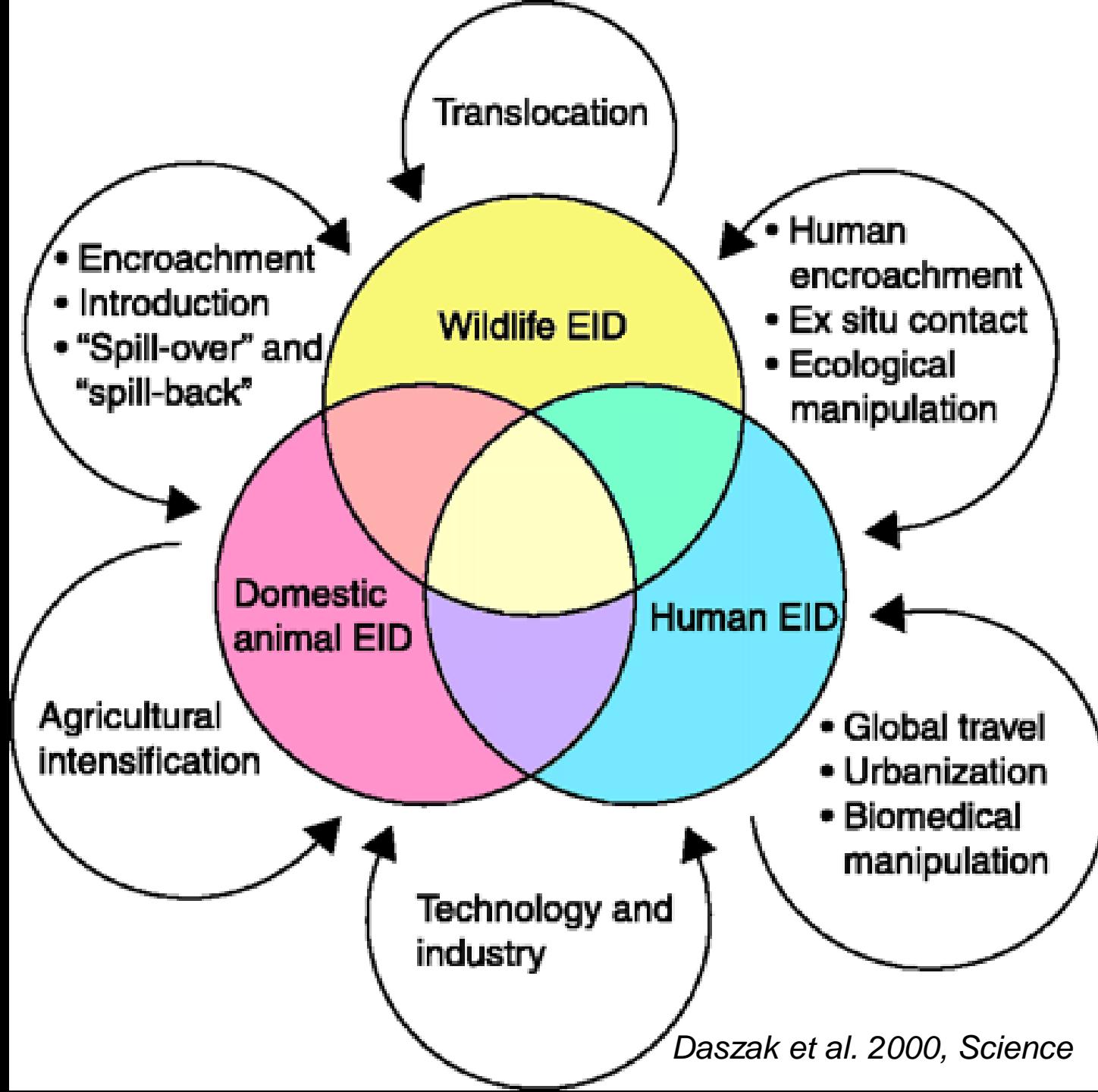




## The paradox:

-Emerging virus infections in the past decade -

(source: WHO)





# Pubmed articles on emerging viruses in wildlife, 2006 to 2010

Virus group	No. of articles	Virus group	No. of articles
Adenovirus	9	Iridoviridae	16
Arenaviridae	9	Morbillivirus	23
Bat viruses	35	Papillomavirus	13
Bornavirus	4	Paramyxoviridae	10
Caliciviridae	9	Parvovirus	5
Circovirus	8	Picornaviridae	9
Coronavirus	9	Polyomavirus	6
Fish viruses	67	Poxvirus	24
Flaviviridae	43	Primate viruses	54
Hantavirus	41	Reoviridae	16
Hepatitis E virus	6	Retroviridae	14
Herpesvirus	33	Rhabdovirus	55
Influenza virus	103	Togaviridae	23



# Important new viral diseases in wildlife, 2006-2010

Virus	Wildlife species	Novelty
West Nile virus	Multiple birds and mammals	New range
Hepatitis E virus	Wild boar	New virus
HPAIV H5N1 virus	Multiple birds and mammals	New hosts
Canine distemper virus	Multiple carnivores	New hosts
Dolphin morbillivirus	Multiple cetaceans	New hosts, re-emergence
Phocine distemper virus	Multiple carnivores	New hosts, re-emergence
African swine fever virus	Wild boar	New range
Bluetongue virus	Multiple mammals	New hosts
Feline leukemia virus	Florida panther, Iberian lynx	New hosts
Border disease virus	Pyrenean chamois	New host
Coronaviruses	Multiple bats	New viruses
Nipah virus	Fruit bat	New range
Ebola virus	Multiple species	Increased incidence
Provent. Dilat. Dis. virus	Psittacines	Etiology determined
Usutu virus	Multiple birds	New range
Amphibian ranaviruses	Multiple amphibians	New viruses

# Elephant endotheliotropic herpes virus

- With few resources, researchers work to contain fatal elephant virus.  
**Am J Vet Res.** 2011 Aug;72(8):1006.
- Nonfatal clinical presentation of elephant endotheliotropic herpes virus discovered in a group of captive Asian elephants (Elephas maximus).  
Schaftenaar W, Reid C, Martina B, Fickel J, Osterhaus AD.  
**J Zoo Wildl Med.** 2010 Dec;41(4):626-32.
- Virus suspect identified in elephant deaths.  
Ferber D.  
**Science.** 1999 Feb 19;283(5405):1093-4.

# Herpesvirus claims another elephant as search for answers continues

With few resources, researchers work to contain fatal elephant virus

R. Scott Nolen, *Am J Vet Res.* 2011 Aug;72(8):1006

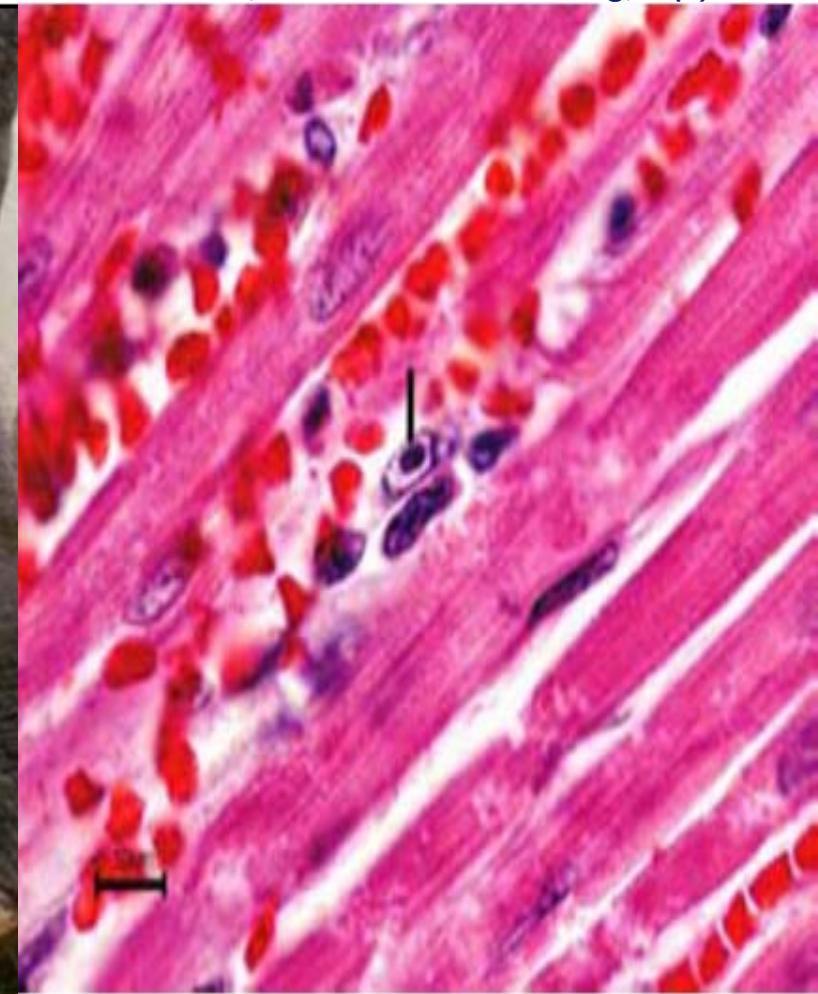
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Elephant endotheliotropic herpesviruses were first identified in 1995 when Kumari (above) died of infection at the National Zoological Park.

Courtesy of Jessie Cohen/Smithsonian National Zoological Park



Viral inclusion bodies in endothelial cells are a hallmark of herpesvirus infection.

Courtesy of Smithsonian National Zoological Park

# Herpesvirus claims another elephant as search for answers continues

With few resources, researchers work to contain fatal elephant virus



Sri Lankan scientists are collaborating with the National Elephant Herpesvirus Laboratory to shed light on the impact EEHVs are having on Asian elephants in the wild.

Courtesy of Amanda Perez/Smithsonian National Zoological Park

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"There are plenty of ugly diseases that kill baby elephants, but this one is terrifying. This is the one we all worry about." —Dr. Ellen Wiedner, who treated one of only eight elephants to survive severe **elephant endotheliotropic herpesvirus** infection

# Nonfatal Clinical Presentation of Elephant Endotheliotropic Herpes Virus Discovered in a Group of Captive Asian Elephants (*Elephas maximus*)

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**Figure 1.** Several elephant endotheliotropic herpes virus-1-related mucosal lesions on the palate of a 36-yr-old female Asian elephant. One lesion is located in a nonpigmented area surrounded by pigmented tissue. Note the orifice of the vomeronasal organ (arrow).



**Figure 2.** Elephant endotheliotropic herpes virus-related vesicles in the vestibulum vaginae of a 36-yr-old female Asian elephant carrying a dead full-grown calf that was 2 mo past due.



## Serology: an indication for viral presence

- Serosurvey for selected infectious disease agents in free-ranging black and white rhinoceros in Africa.  
Fischer-Tenhagen C, Hamblin C, Quandt S, Frölich K
- Canine distemper virus antibodies in the Asian elephant (*Elaphas maximus*).  
Oni O, Wajjwalku W, Boodde O, Chumsing W.  
Vet Rec. 2006 Sep 23;159(13):420-1.
- Antibodies against some viruses of domestic animals in southern African wild animals.  
Barnard BJ.  
Onderstepoort J Vet Res. 1997 Jun;64(2):95-110.
- Enzyme-linked immunosorbent assays for the detection of antibody to Crimean-Congo haemorrhagic fever virus in the sera of livestock and wild vertebrates.  
Burt FJ, Swanepoel R, Braack LE.  
Epidemiol Infect. 1993 Dec;111(3):547-57.
- Serological evidence of herpesvirus infection in captive Asian elephants (*Elephas maximus*).  
Metzler AE, Ossent P, Guscetti F, Rübel A, Lang EM.  
J Wildl Dis. 1990 Jan;26(1):41-9

# Serosurvey for selected infectious disease agents in free-ranging black and white rhinoceros in Africa.

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Fischer-Tenhagen C, Hamblin C, Quandt S, Frölich K

Two hundred and eighty one serum samples collected from free-ranging black (*Dicerosbicornis*) and white (*Ceratotherium simum*) rhinoceros, in the Republic of South Africa (RSA), Namibia, and Kenya from 1987–97, were examined for antibody to 16 different infectious agents.

Positive antibody titers were detected against Akabane (59.8%), bluetongue (55%), African horse sickness (27.9%), epizootic haemorrhagic disease of deer (19.4%), parainfluenza type 3 (25.3%), bovine herpes virus 1 (3.1%), equine herpes virus 1 (8.8%) and bovine viral diarrhea (1.2%) viruses, and four serovars of *Leptospira interrogans*, (ranging 1.2 to 8.8%).

Interspecies differences were detected for African horse sickness, epizootic haemorrhagic disease of deer and parainfluenza type 3 viruses.

There appeared to be some geographic variation in the prevalence of antibody for African horse sickness, bluetongue, epizootic haemorrhagic disease of deer, parainfluenza type 3, equine herpes virus 1 and *Leptospira interrogans* serovar *bratislava*.



# Endogenous viruses in elephants



## Endogenous non-retroviral RNA virus elements in mammalian genomes.

Horie M, Honda T, Suzuki Y, Kobayashi Y, Daito T, Oshida T, Ikuta K, Jern P, Gojobori T, Coffin JM, Tomonaga K.

Nature. 2010 Jan 7;463(7277):84-7.



## Discovery and characterization of mammalian endogenous parvoviruses.

Kapoor A, Simmonds P, Lipkin WI.

J Virol. 2010 Dec;84(24):12628-35. Epub 2010 Oct 13.

## Characterization of an endogenous retrovirus class in elephants and their relatives.

Greenwood AD, Englbrecht CC, MacPhee RD.

BMC Evol Biol. 2004 Oct 11;4:38.

## Evolution of endogenous retrovirus-like elements of the woolly mammoth (*Mammuthus primigenius*) and its relatives.

Greenwood AD, Lee F, Capelli C, DeSalle R, Tikhonov A, Marx PA, MacPhee RD.

Mol Biol Evol. 2001 May;18(5):840-7.

# Identification of new viral pathogens

- 1995 CDV as the cause of mass mortality in Serengeti lions
- 1996  $\gamma$ -herpesvirus in seals (phocid herpesvirus-2)
- 1997 monk seal morbilliviruses (MSMV-WA/G)
- 1997 influenza A (H5N1) virus in humans
- 1998 lentivirus from Talapoin monkeys (SIVtal)
- 1999 influenza B virus in seals
- 2000 human metapneumovirus (hMPV)
- 2002 re-emerging PDV in Europe
- 2003 SARS CoV cause of SARS in humans (Koch's postulates)
- 2003 influenza A (H7N7) virus in humans
- 2004 fourth human coronavirus (CoV NL)
- 2005 H16 influenza A viruses (new HA!) in black headed gulls
- 2008 dolphin herpesvirus
- 2009 deer astrovirus
- 2010 human astrovirus, human picobirnavirus
- 2011 ferret coronavirus, porcine picobirnavirus, stone marten anellovirus.  
influenza A (H1N1) virus in dogs...

# Identification of viral pathogens (1/2)

- 1977 non-mammalian papillomavirus: Finch papillomavirus
- 1984 phocid herpesvirus (phocid alpha herpesvirus-1, PhHV-1)
- 1985 phocid morbillivirus (phocid distemper virus, PDV)
- 1988 picornavirus in seals (affected by PDV infection)
- 1990 orthopoxvirus in seals (affected by PDV infection)
- 1992 porpoise and dolphin morbilliviruses causing mortality
- 1992 rhabdovirus of dolphins





# Poxvirus Taxonomy

## Genus

avipoxvirus

capripoxvirus

leporipoxvirus

molluscipoxvirus

orthopoxvirus

parapoxvirus

suispoxvirus

yatapoxvirus

## Species

camelpox virus

cowpox virus

ectromelia virus

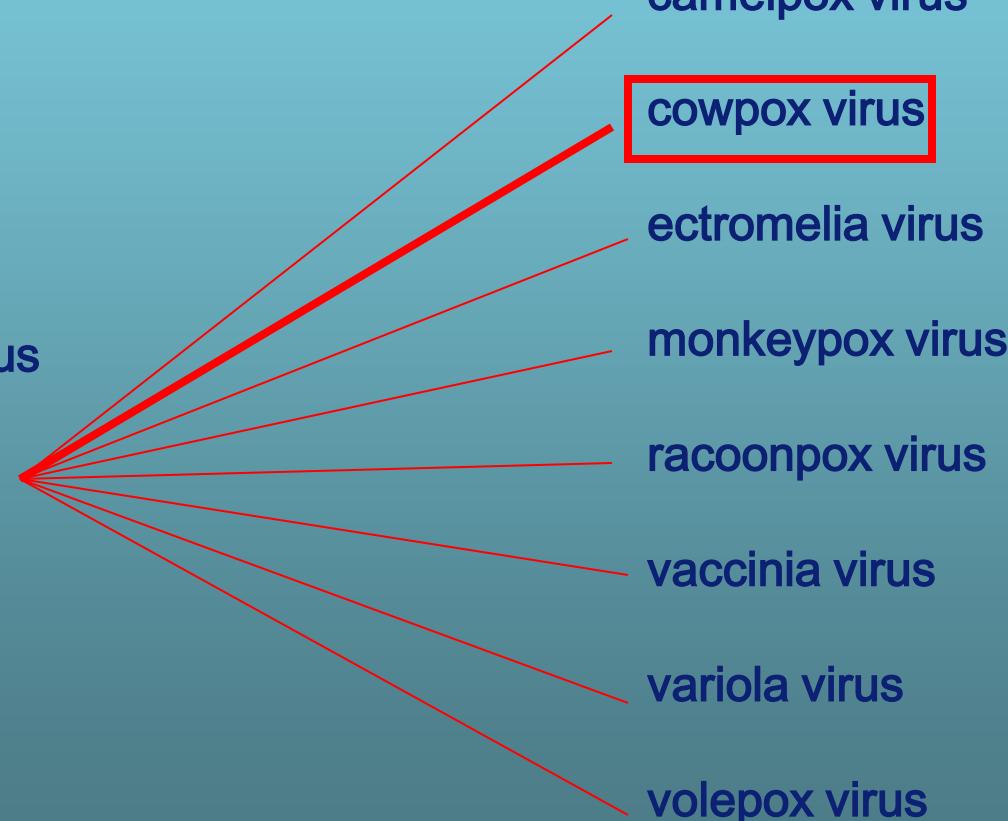
monkeypox virus

raccoonpox virus

vaccinia virus

variola virus

volepox virus



cowpox



Wolfs et al. E.I.D. 2002

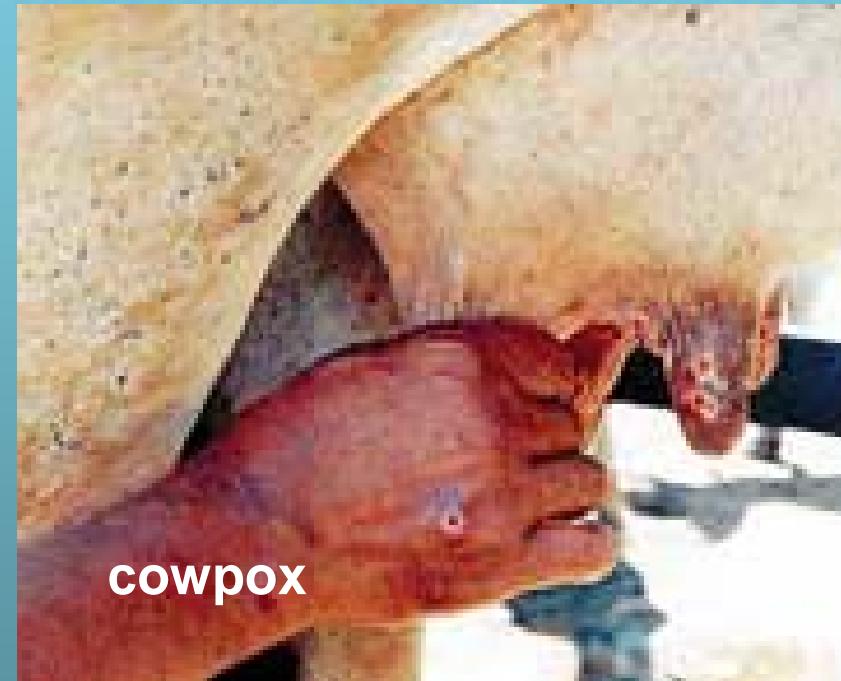


Figure 1. African child with disseminated monkeypox. Note postauricular adenopathy (courtesy of Leo Lanoie, Prince Albert Parkland Health Region, Saskatchewan, Canada).

# Since eradication smallpox: more animal poxvirus infections in humans?

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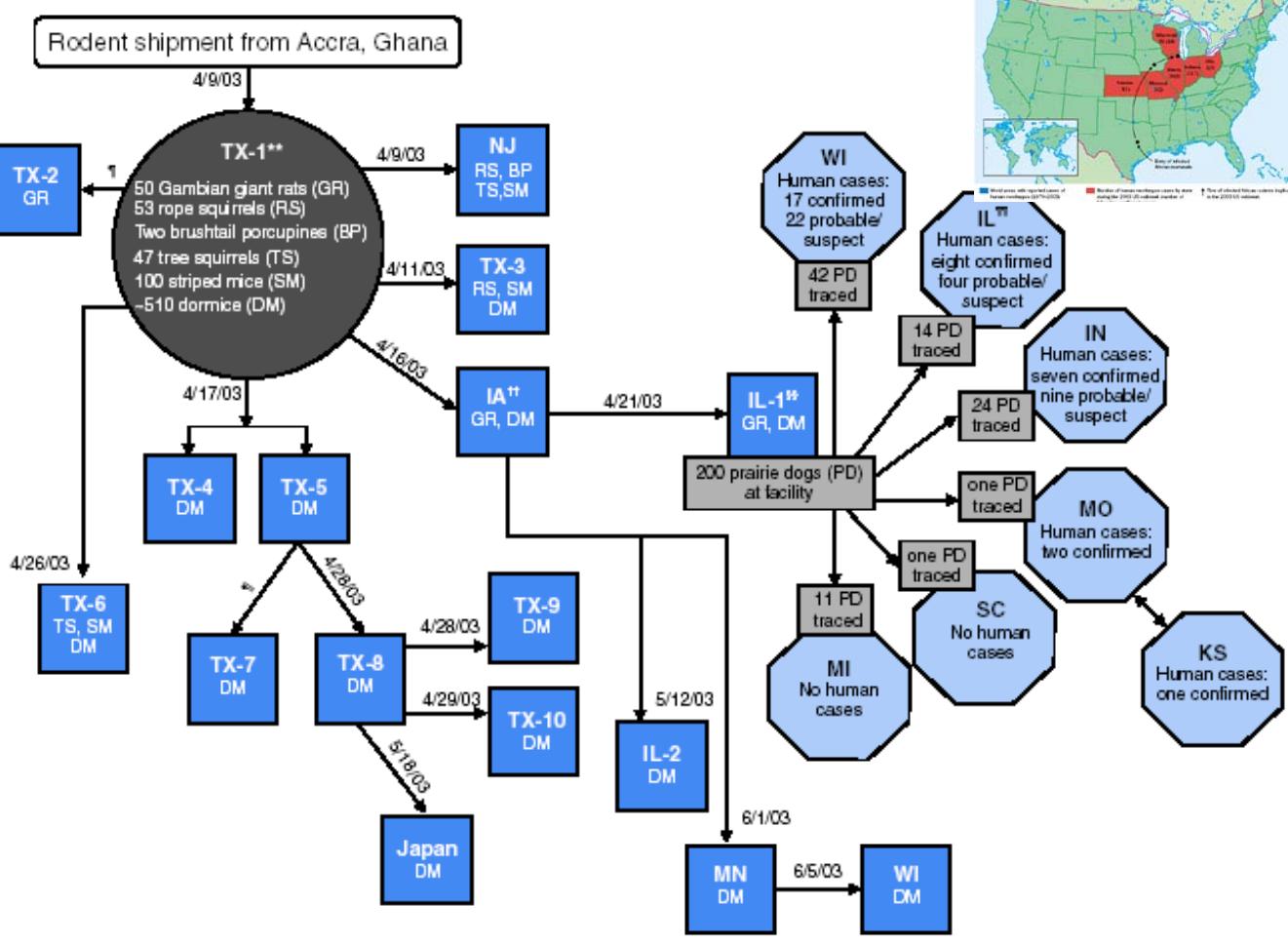
ProMED-mail 9 Jan 2003

Stittelaar et al., Nature, 2006

Pelkonen et al. E.I.D. 2003; 9:1458-1461

# Multistate outbreak of monkeypox

## Illinois, Indiana, and Wisconsin, 2003





# Cowpoxvirus infections in elephants

## Human cowpox virus infection acquired from a circus elephant in Germany.

Hemmer CJ, Littmann M, Löbermann M, Meyer H, Petschaelis A, Reisinger EC.

Int J Infect Dis. 2010 Sep;14 Suppl 3:e338-40.

## Rat-to-elephant-to-human transmission of cowpox virus.

Kurth A, Wibbelt G, Gerber HP, Petschaelis A, Pauli G, Nitsche A. Emerg Infect Dis. 2008 Apr;14(4):670-1.

## Cowpox virus infection causing stillbirth in an Asian elephant (Elphas maximus).

Wisser J, Pilaski J, Strauss G, Meyer H, Burck G, Truyen U, Rudolph M, Frölich K.

Vet Rec. 2001 Aug 25;149(8):244-6.

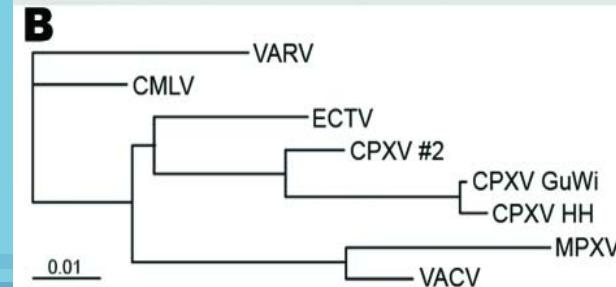
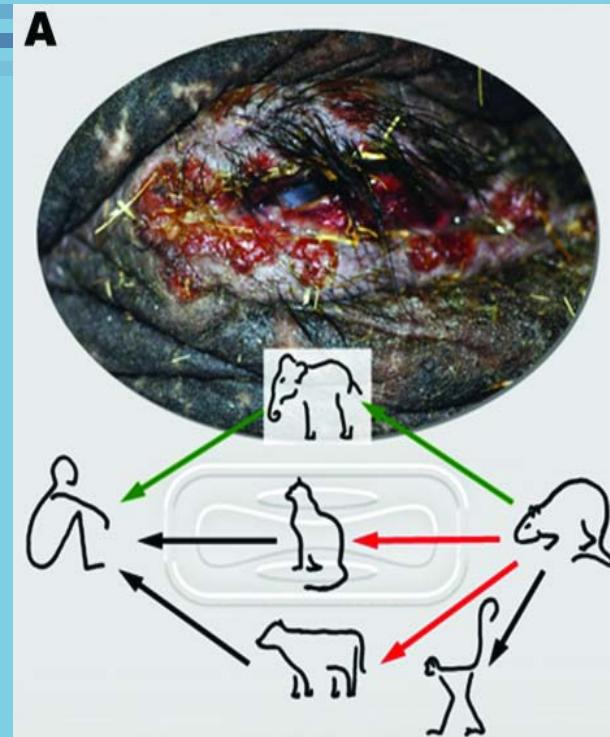
## Characterization of orthopoxviruses isolated from man and animals in Germany.

Meyer H, Schay C, Mahnel H, Pfeffer M. Arch Virol. 1999;144(3):491-501.

## A method for serological differentiation of closely related poxviruses.

Maltseva NN, Marennikova SS.

Acta Virol. 1976 Jun;20(3):250-52.



# Human cowpox virus infection acquired from a circus elephant in Germany



**Figure 1.** Clinical evolution of the cowpox lesion over time (days after the onset of symptoms).

# Characterization of orthopoxviruses isolated from man and animals in Germany

H. Meyer<sup>1</sup>, C. Schay<sup>1</sup>, H. Mahnel<sup>2</sup>, and M. Pfeffer<sup>2</sup>

Fourteen orthopoxvirus strains isolated from humans, cats, a dog, a cow, and an elephant in Germany were characterized.

All were classified as cowpox virus based on haemorrhagic lesions induced on the Chorioallantoic membrane of chicken eggs and reactivity of a 160 kDa protein with anti-A-type inclusion protein hyperimmun serum in a Western blot. More detailed Comparison of the isolates by restriction endonuclease mapping using *Hind*III and *Xba*I demonstrated a close relationship between all isolates and confirmed them as cowpox viruses.

One group consisting of five closely related isolates contained a unique 4.0 kb *Hind*III fragment.

In a Southern blot this fragment failed to hybridize with other cowpox virus isolates including the reference strain.



Fig. 3. Map of Germany depicting where the 14 cowpox virus strains described in this study had been isolated

# Poxvirus in a rhino

- Characterization of a fowlpox virus isolated from a rhinoceros].

Mayr A, Mahnel H.

Arch Gesamte Virusforsch. 1970;31(1):51-60.





## EMC virus in elephants

*Cardiovirus* is a genus within the family *Picornaviridae*

- An outbreak of encephalomyocarditis-virus infection in free-ranging African elephants in the Kruger National Park.

Grobler DG, Raath JP, Braack LE, Keet DF, Gerdes GH, Barnard BJ, Kriek NP, Jardine J, Swanepoel R.

Onderstepoort J Vet Res. 1995 Jun;62(2):97-108

- Encephalomyocarditis virus infection of captive elephants.

Simpson CF, Lewis AL, Gaskin JM.

J Am Vet Med Assoc. 1977



# EMC vaccination elephants

Hunter et al., Vaccine 1998



The efficacy of an experimental oil-adjuvanted encephalomyocarditis vaccine in elephants, mice and pigs.

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- An oil-adjuvanted inactivated encephalomyocarditis (EMC) vaccine was developed.
- Mice showed protection against challenge and pigs developed high antibody levels.
- Vaccinated elephants developed high antibody titers which protected all vaccines from a challenge roughly two months post-vaccination, whereas controls developed fatal or sub-clinical myocarditis.
- Ethics?



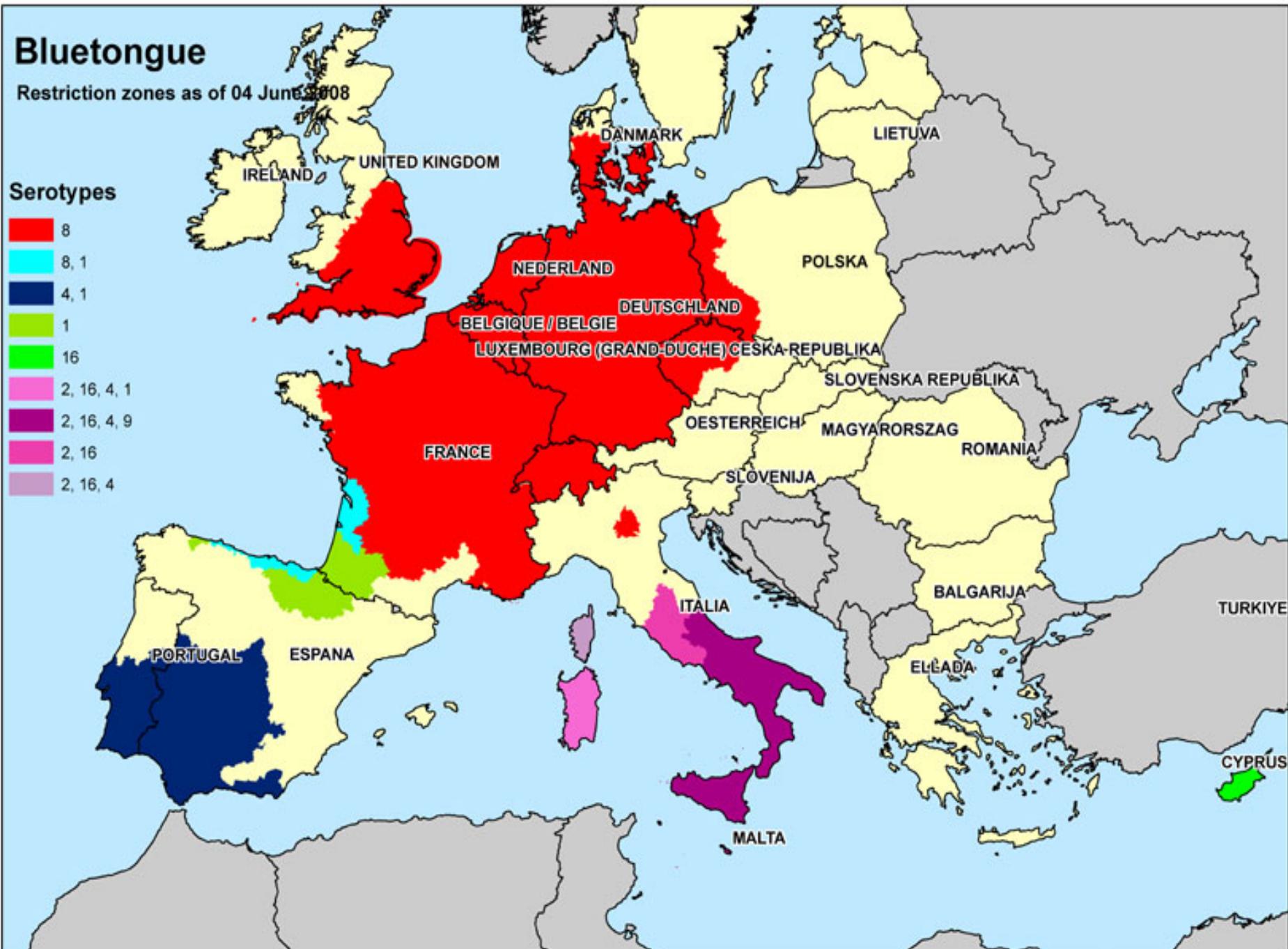
Figure 1 The heart function of the experimental elephants was monitored using an ECG after challenge with the EMC virus.

# Bluetongue

Restriction zones as of 04 June 2008

## Serotypes

- 8
- 8, 1
- 4, 1
- 1
- 16
- 2, 16, 4, 1
- 2, 16, 4, 9
- 2, 16
- 2, 16, 4



# Bluetongue antibodies in wildlife Africa

Epidemiologic study of bluetongue in sheep, cattle and different species of wild animals in the Ivory Coast.

Formenty P, Domenech J, Lauginie F, Ouattara M, Diawara S, Raath JP, Grobler D, Leforban Y, Angba A.

Rev Sci Tech. 1994 Sep;13(3):737-51.

antibodies against BTV in:

kob (Kobus kob)

common waterbuck (Kobus ellipsiprymnus)

roan antelope (Hippotragus equinus),

buffalo (Syncerus caffer)

hartebeest (Alcelaphus buselaphus)

elephant (Loxodonta africana)



©IAH 2006

# African horse sickness: elephants as a reservoir?

- Epidemiology of African horsesickness: antibodies in free-living elephants (*Loxodonta africans*) and their response to experimental infection.  
Barnard BJ, Bengis RG, Keet DF, Dekker EH.  
Onderstepoort J Vet Res. 1995 Dec;62(4):271-5.
- An attempt to define the host range for African horse sickness virus (Orbivirus, Reoviridae) in east Africa, by a serological survey in some Equidae, Camelidae, Loxodontidae and Carnivores  
Binepal VS, Wariru BN, Davies FG, Soi R, Olubayo R.  
Vet Microbiol. 1992 Apr;31(1):19-23
- Elephants and zebras as possible reservoir hosts for African horse sickness virus.  
Davies FG, Otieno S.  
Vet Rec. 1977 Apr 2;100(14):291-2.

## FMDV in elephants

- Foot and mouth disease in elephant (*Elephas maximus*).  
Rahman H, Dutta PK, Dewan JN.  
Zentralbl Veterinarmed B. 1988 Jan;35(1):70-1.
- An outbreak of foot-and-mouth disease in Indian elephants (*Elephas maximus*).  
Pyakural S, Singh U, Singh NB.  
Vet Rec. 1976 Jul 10;99(2):28-9.



# Morbilliviruses crossing species barriers

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PDV in European Harbour seals  
Nature 1988 / Science 2002



CDV in Baikal seals  
Nature 1988



CDV in Caspian seals  
EID 2000



CDV in Serengeti lions  
Vaccine 1994



DMV in Med. monk seals  
Nature 1997



CDV in macaques  
China, EID 2011

should we continue measles vaccination for ever?

# CDV antibodies... what about disease?

- Canine distemper virus antibodies in the Asian elephant (*Elphas maximus*).

Oni O, Wajjwalku W, Boodde O, Chumsing W.

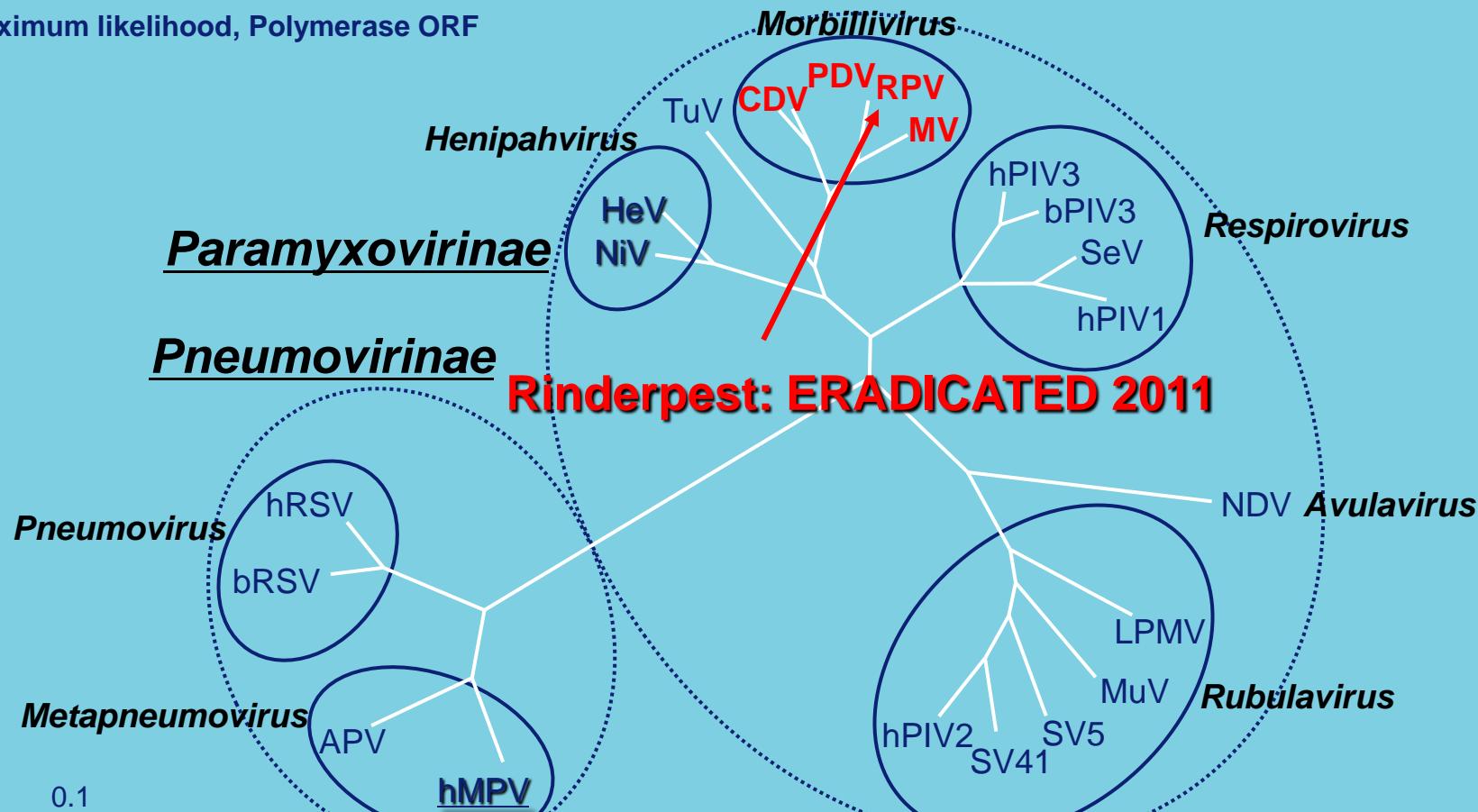
Vet Rec. 2006 Sep 23;159(13):420-1.





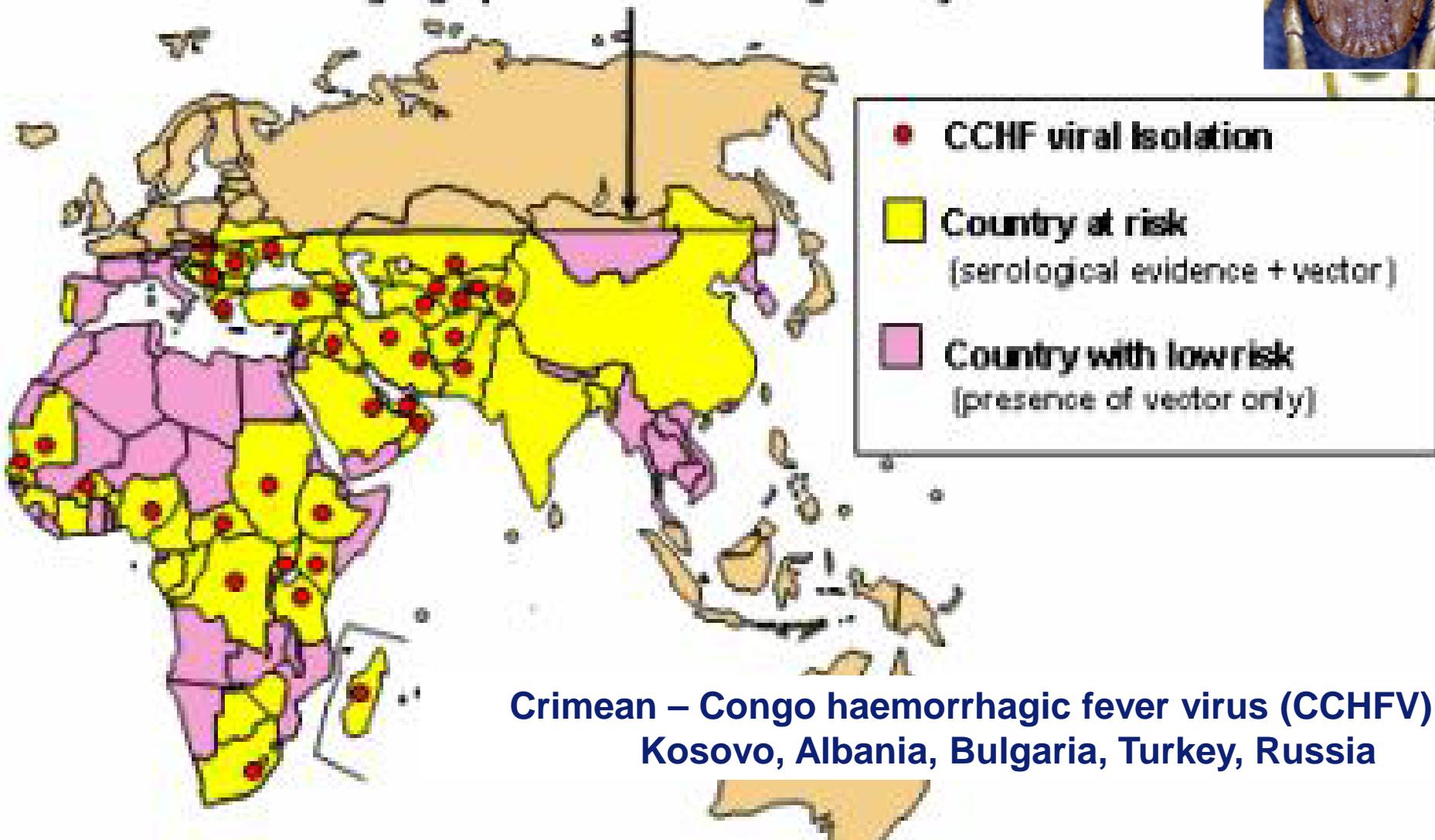
## Morbilliviruses: a continuing story!!!

DNA Maximum likelihood, Polymerase ORF



# CCHF: geographic distribution

North limit for the geographic distribution of genus *Hyalomma* ticks



## CCHV antibodies in African elephants

- Enzyme-linked immunosorbent assays for the detection of antibody to Crimean-Congo haemorrhagic fever virus in the sera of livestock and wild vertebrates.

Burt FJ, Swanepoel R, Braack LE.

**Epidemiol Infect.** 1993 Dec;111(3):547-57.

The CELISA was applied to the sera of 960 wild vertebrates from a nature reserve in South Africa, and the prevalence of antibody was found to be greatest in large mammals such as rhinoceros, giraffe and buffalo, which are known to be the preferred hosts of the adult tick (Hyalomma) vectors of the virus.

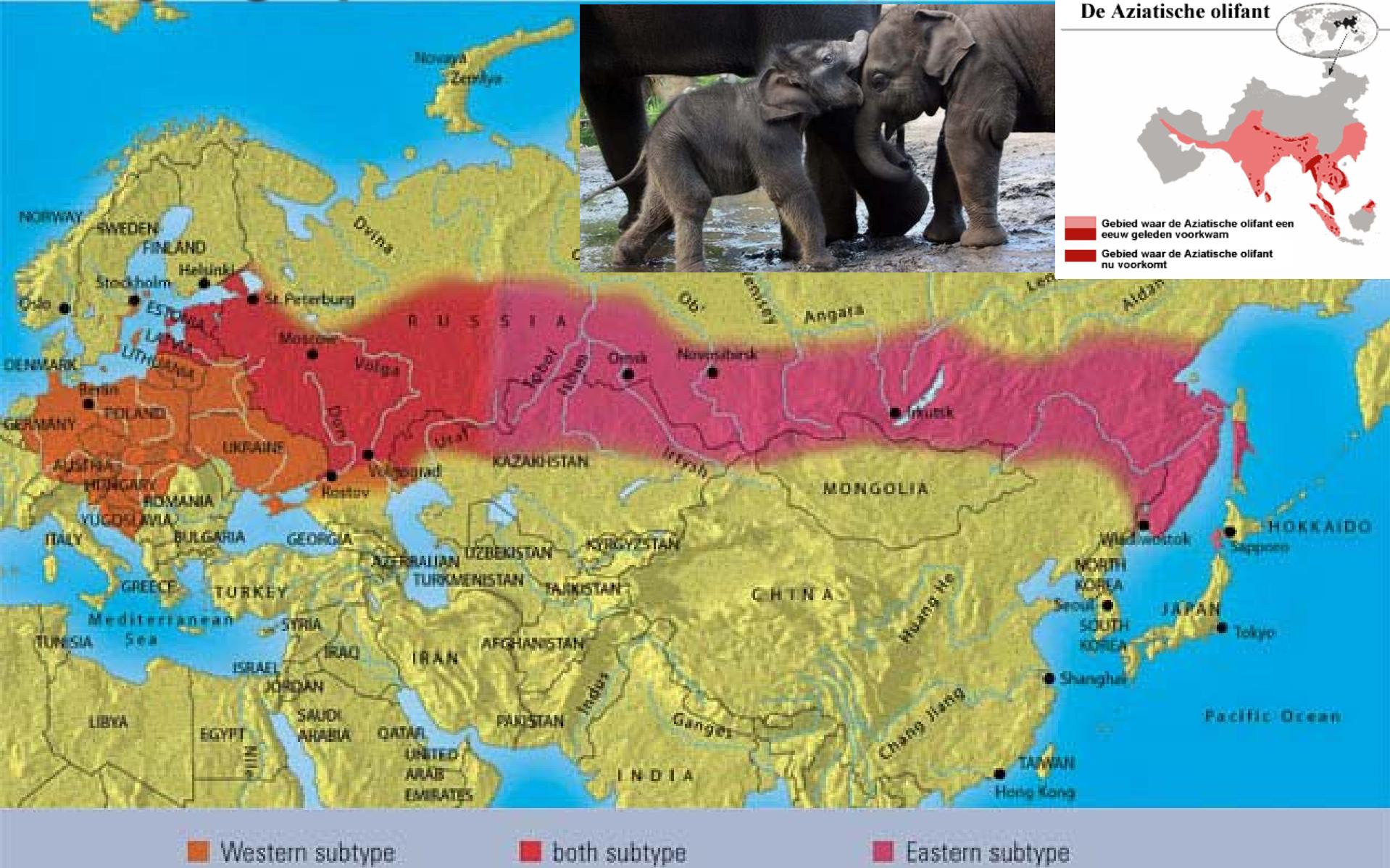
# TICK-BORNE ENCEPHALITIS

## -geographical distribution-

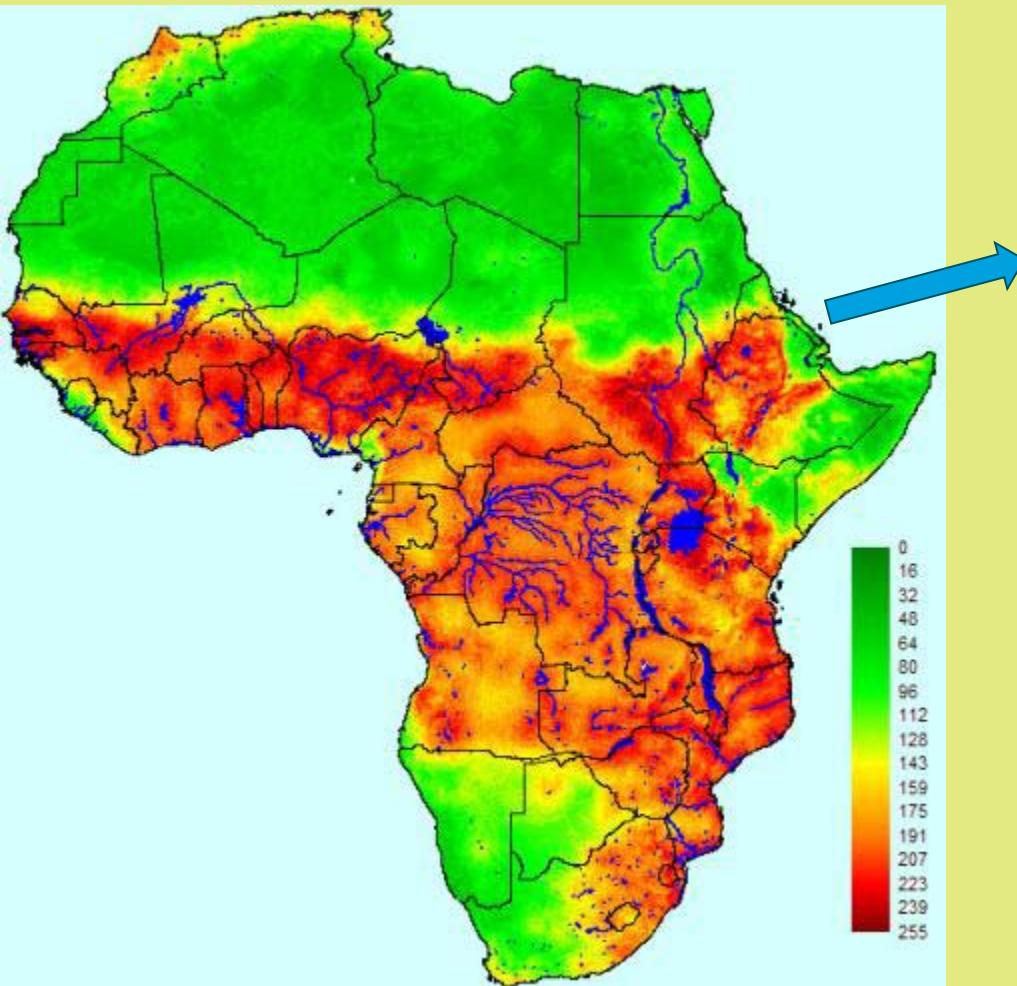
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De Aziatische olifant



## Climate effects on mosquito-borne infections



RVF outbreaks: massive mortality ungulates (and HF in humans)

Food shortage, increased consumption of bush meat

Increased risk of zoonotic infections (eg monkeypox)

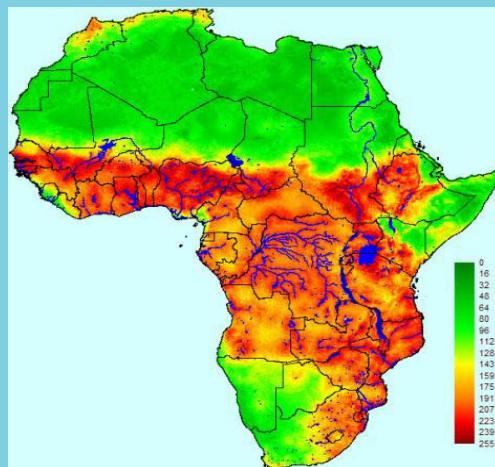
**Rift Valley Fever (RVF)** is a viral zoonosis (affects primarily domestic livestock, but can be passed to humans) causing fever. It is spread by the bite of infected mosquitoes, typically the Aedes or Culex genera.

# Rift valley fever antibodies in elephants

Prevalence of antibodies against Rift Valley fever virus in Kenyan wildlife.

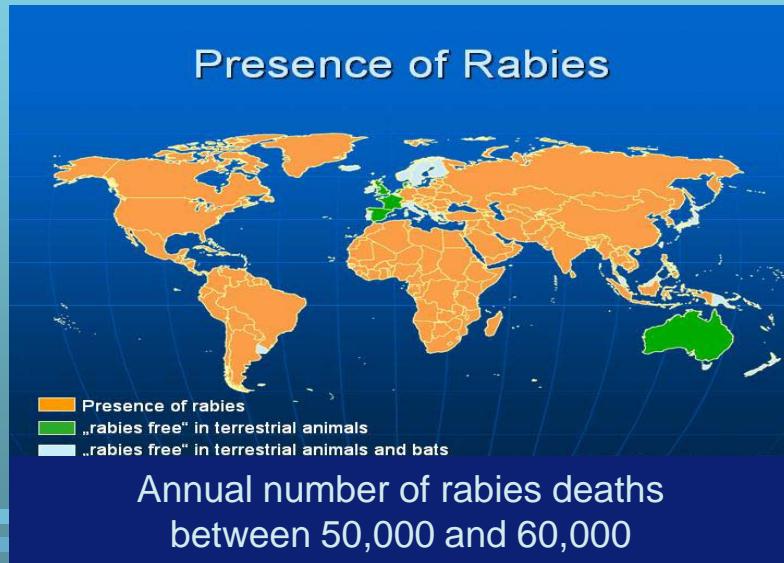
Evans A, Gakuya F, Paweska JT, Rostal M, Akoolo L, Van Vuren PJ, Manyibe T, Macharia JM, Ksiazek TG, Feikin DR, Breiman RF, Kariuki Njenga M.

**Epidemiol Infect.** 2008 Sep;136(9):1261-9. Epub 2007 Nov 8



# Duvenhage (rabies) virus infection in The Netherlands: “out of Africa”

failure of treatment  
of a patient in  
The Netherlands!



- van Thiel et al., Euro surveill., 2008
- van Thiel et al., Plos Negl. Trop. Dis. 2009

# Rabies in elephants

- Serum antibody titers following routine rabies vaccination in African elephants.  
Miller MA, Olea-Popelka F.  
J Am Vet Med Assoc. 2009 Oct 15;235(8):978-81.
- Results of vaccination of Asian elephants (*Elephas maximus*) with monovalent inactivated rabies vaccine.  
Isaza R, Davis RD, Moore SM, Briggs DJ.  
Am J Vet Res. 2006 Nov;67(11):1934-6.
- First reported case of elephant rabies in Sri Lanka.  
Wimalaratne O, Kodikara DS.  
Vet Rec. 1999 Jan 23;144(4):98.

# CONCLUSIONS:

## VIRUSES in elephants and rhinos

### Points to consider:

- **problems in captivity (pox, herpes, EMC...)**
- **problems in the wild (herpes, EMC, rabies, FMDV...)**
- **reservoirs (BT, AHS, EMC, FMDV...)**
- **virus discovery (classical / advanced techniques)**
- **importance for translocation / conservation**
- **surveillance (syndromic, sampling / laboratory)**
- **prevention (wildlife- / captive management...)**

