

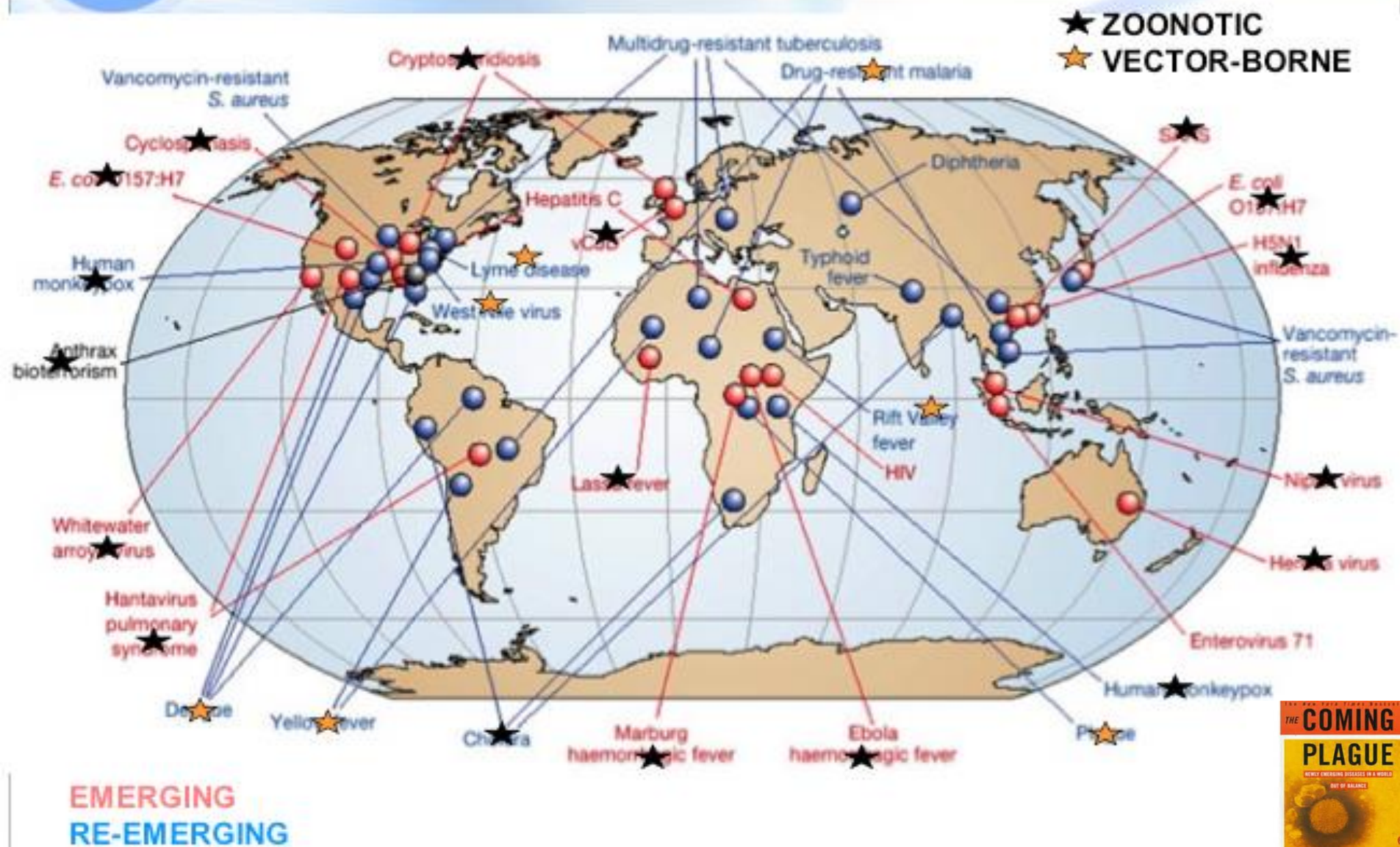
BATS:

rabies and other infectious diseases

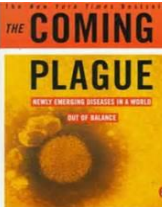


CE Rupprecht VMD, MS, PhD
CEO, LYSSA LLC;
WHO Expert Technical Advisor on Rabies;
RITA INC, International Steering Committee;
Scientific Advisory Board, Bat Conservation
International

Global Emerging Diseases*



* Modified from Morens et al. 2004 Nature 430:242



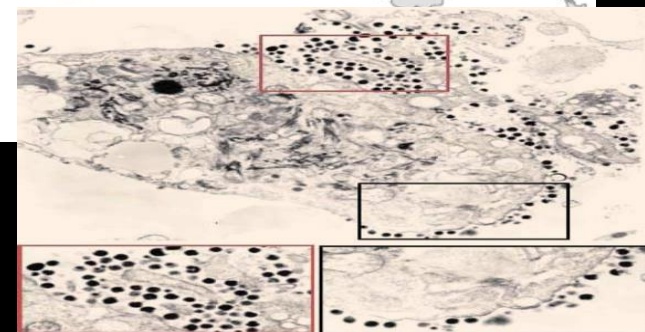
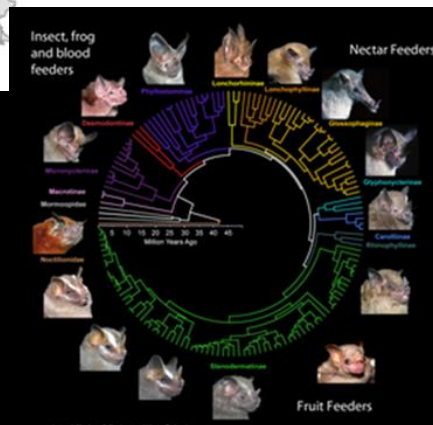
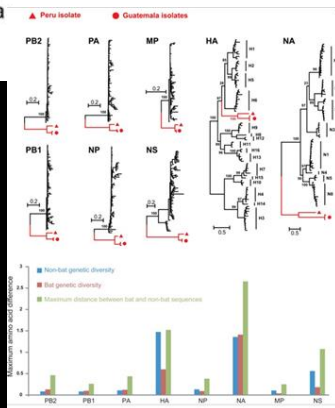
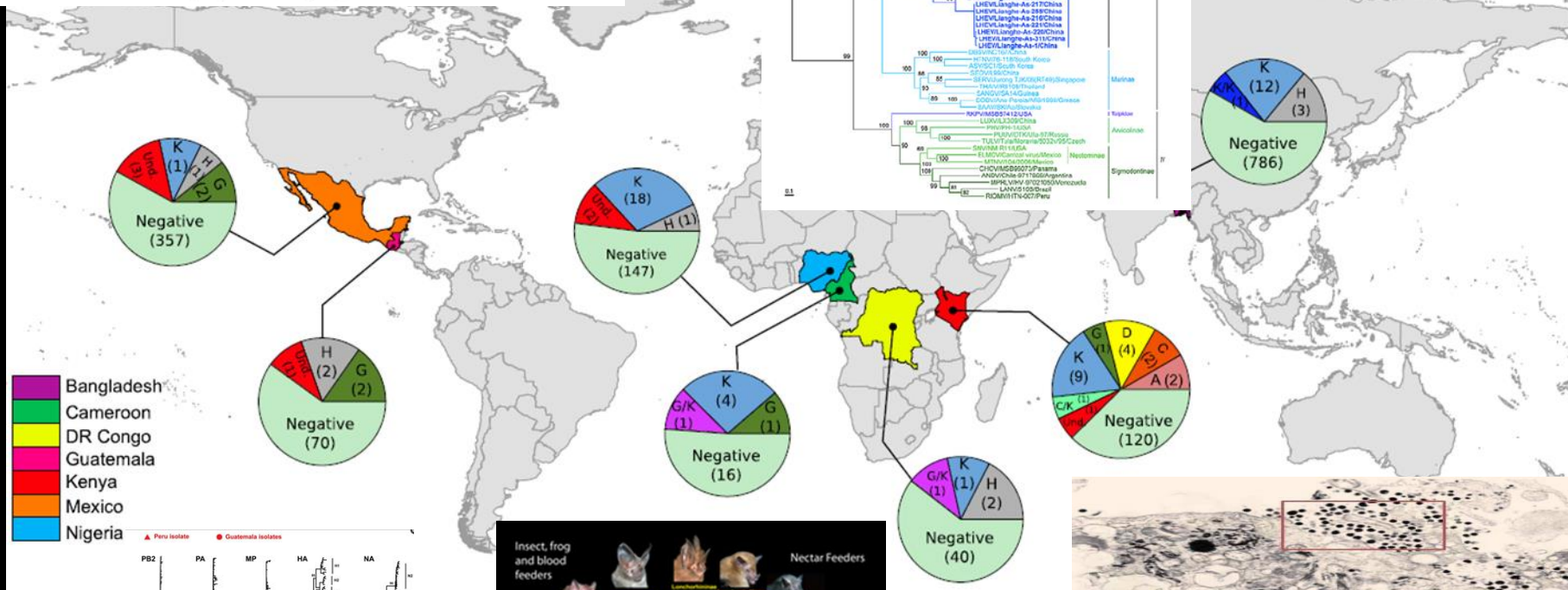
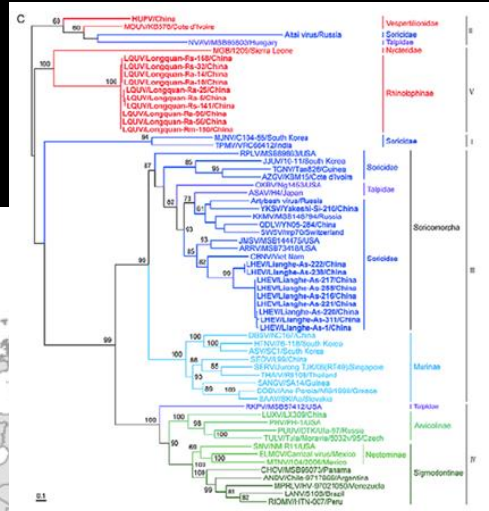
Laurie Garrett
World of a 19th-century doctor
Reporting on the Dark Side

OUTLINE

- Compare basic biological attributes of bats
- Cite selected reports on bat pathogen discovery
- Infer applications from bat lyssavirus introspection as to perceived relevance for other pathogens
- Discuss perspectives on recent agent emergence, host changes, potential management & future concerns

Orthohepadnavirus

Avihepadnavirus



RS. Shabman et al. Isolation and Characterization of a Novel Gammaherpesvirus from a Microbat Cell Line. mSphere, published online February 17, 2016

BASIC BAT ATTRIBUTES THAT MAY BE SUPPORTIVE OF PARASITE/PATHOGEN RICHNESS

Order Chiroptera

Ancient Taxa

Global Distribution

>1,100 species

Volant

Biodiversity

Vagility

Gregariousness

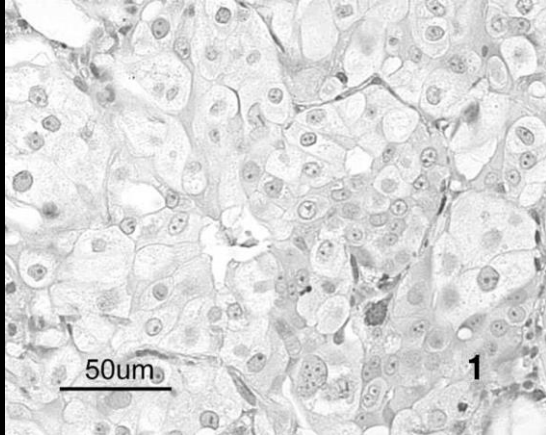
Torpor & Hibernation

High Reproductive Success

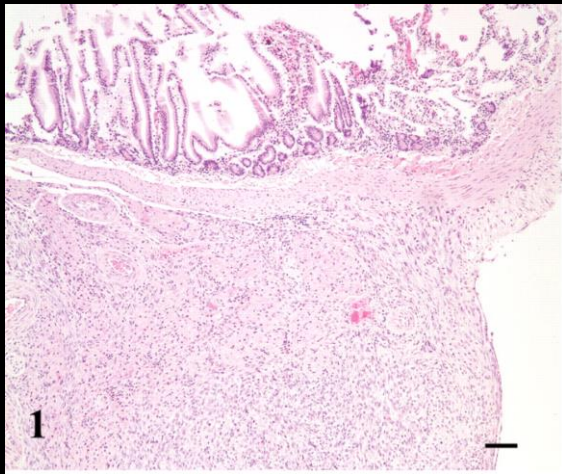
Longevity



REPORTS OF NEOPLASIA IN THE CHIROPTERA ARE RARE



McLelland et al. J VET Diagn Invest 2009;21:160



Bradford et al. J VET Diagn Invest 2010;22:462-465

Olds JE, et al. Retrospective evaluation of neoplasia in a captive population of Egyptian fruit bats (*R. aegyptiacus*). J Zoo Wildl Med. 2015;46:325-32.

Buckles EL. Chiroptera (bats). In: Fowler ME, Miller RE (eds.). Zoo and wildlife medicine, Volume 8. St. Louis (MO): Elsevier; 2014. p. 285–286.

Cushing AC, et al. **Metastatic pancreatic and bronchioloalveolar adenomas** in an Egyptian fruit bat (*Rousettus aegyptiacus*). J Zoo Wildl Med. 2013;44:794–798.

Bradford C, et al. **Gastrointestinal leiomyosarcoma** in an Egyptian fruit bat (*Rousettus aegyptiacus*). J Vet Diagn Invest. 2010;22:462–465.

McLellund DJ, et al. **Sarcomatoid carcinoma** in the lung of an Egyptian fruit bat (*Rousettus aegyptiacus*). J Vet Diagn Invest. 2009;21:160–163.

Siegel-Willot J, et al. Microchip-associated **leiomyosarcoma** in an Egyptian fruit bat (*Rousettus aegyptiacus*). J Zoo Wildl Med. 2007;38:352–356.

McKnight CA, et al. **Papillomavirus-associated basosquamous carcinoma** in an Egyptian fruit bat (*Rousettus aegyptiacus*). J Zoo Wildl Med. 2006;37:193–196.

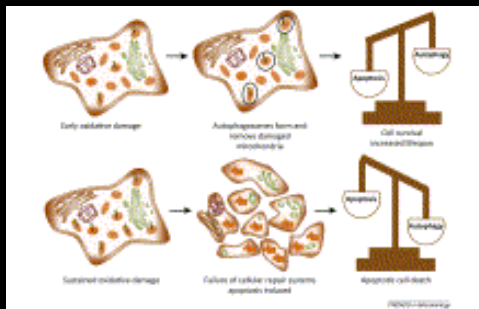
Andreasen C, Dulmstra JR. **Multicentric malignant lymphoma** in a pallid bat. J Wildl Dis. 1996;32:545–547.

Beck M, et al. **Bile duct adenocarcinoma** in a pallid bat (*A. pallidus*). J Wildl Dis. 1982;18:365–367.

Conrad LG, et al. **Cutaneous leiomyosarcoma** in a long-eared bat, *Plecotus townsendii*. J Natl Cancer Inst. 1965;35:95-101.

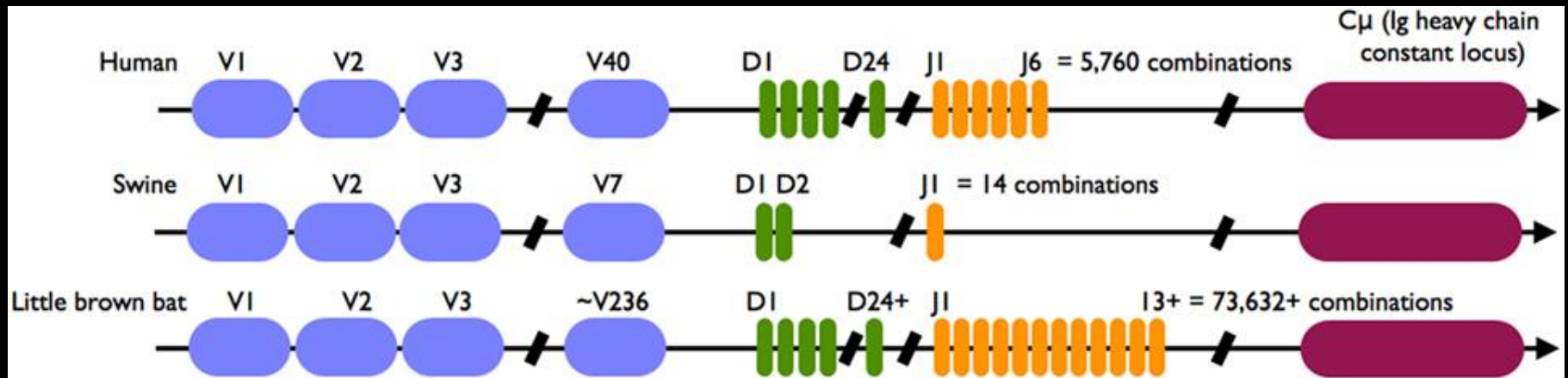
BAT PHYSIOLOGY– “Flight As Fever”?

- Bats experience morbidity to many extracellular infections.
- Bats may control intracellular pathogens via cellular pathways to apoptosis/autophagy.
- These mitigation pathways could support longevity and tumor avoidance.
- Extracellular pathogen-associated morbidity may result from immunopathology.
- Mitochondrial mechanisms of bat physiology may have evolved to mitigate oxidative stress incurred during metabolically costly and ‘hot’ activities such as flight.



Brook & Dobson 2015:23, p172–180, Trends Microbiol
O’Shea et al 2014:20, p.741-745, EID

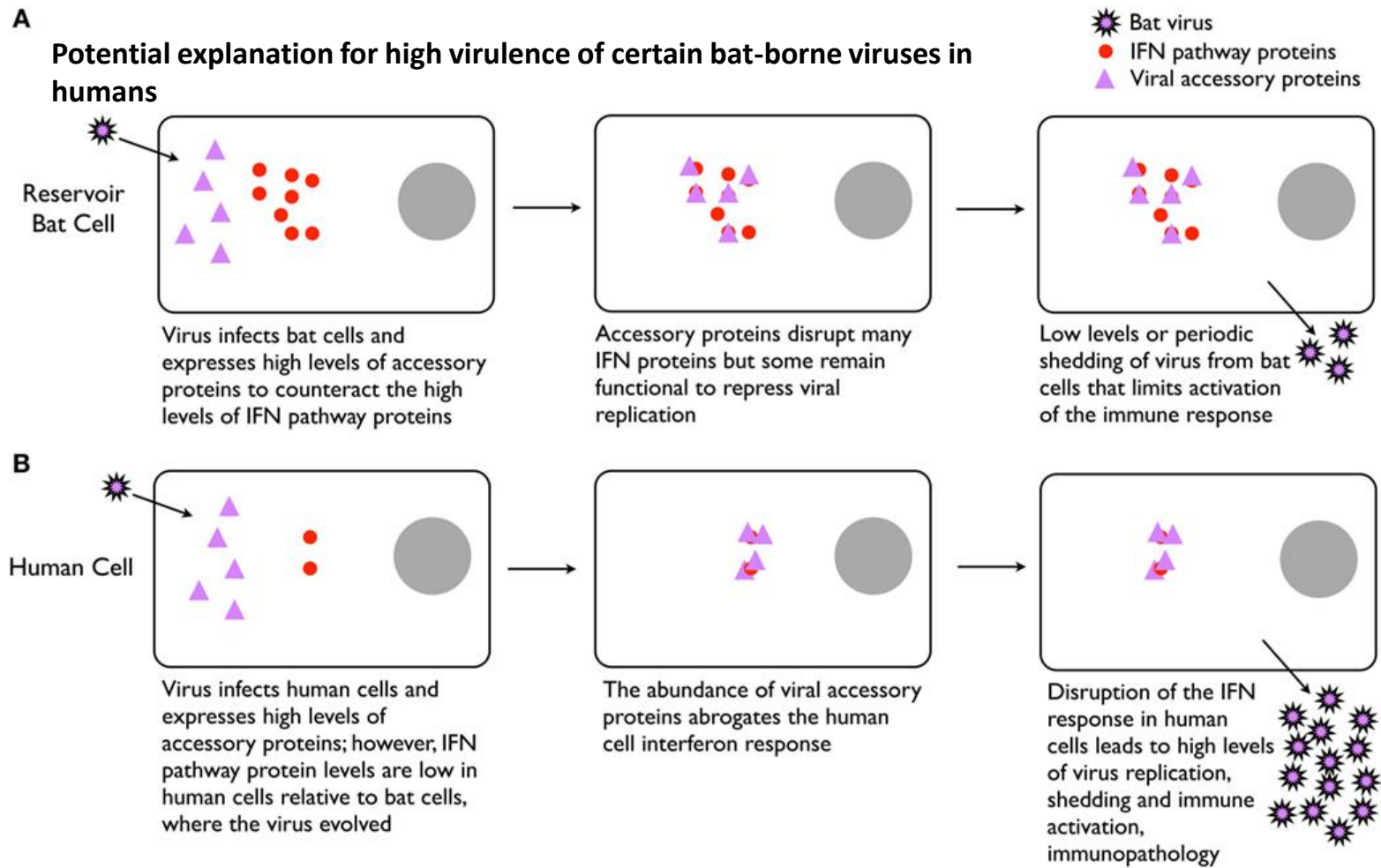
Immunoglobulin combinatorial diversity potential of humans, swine and bats



Schountz T, et al. Immunological Control of Viral Infections in Bats and the Emergence of Viruses Highly Pathogenic to Humans. *Front Immunol.* 2017 Sep 11;8:1098.

A

Potential explanation for high virulence of certain bat-borne viruses in humans



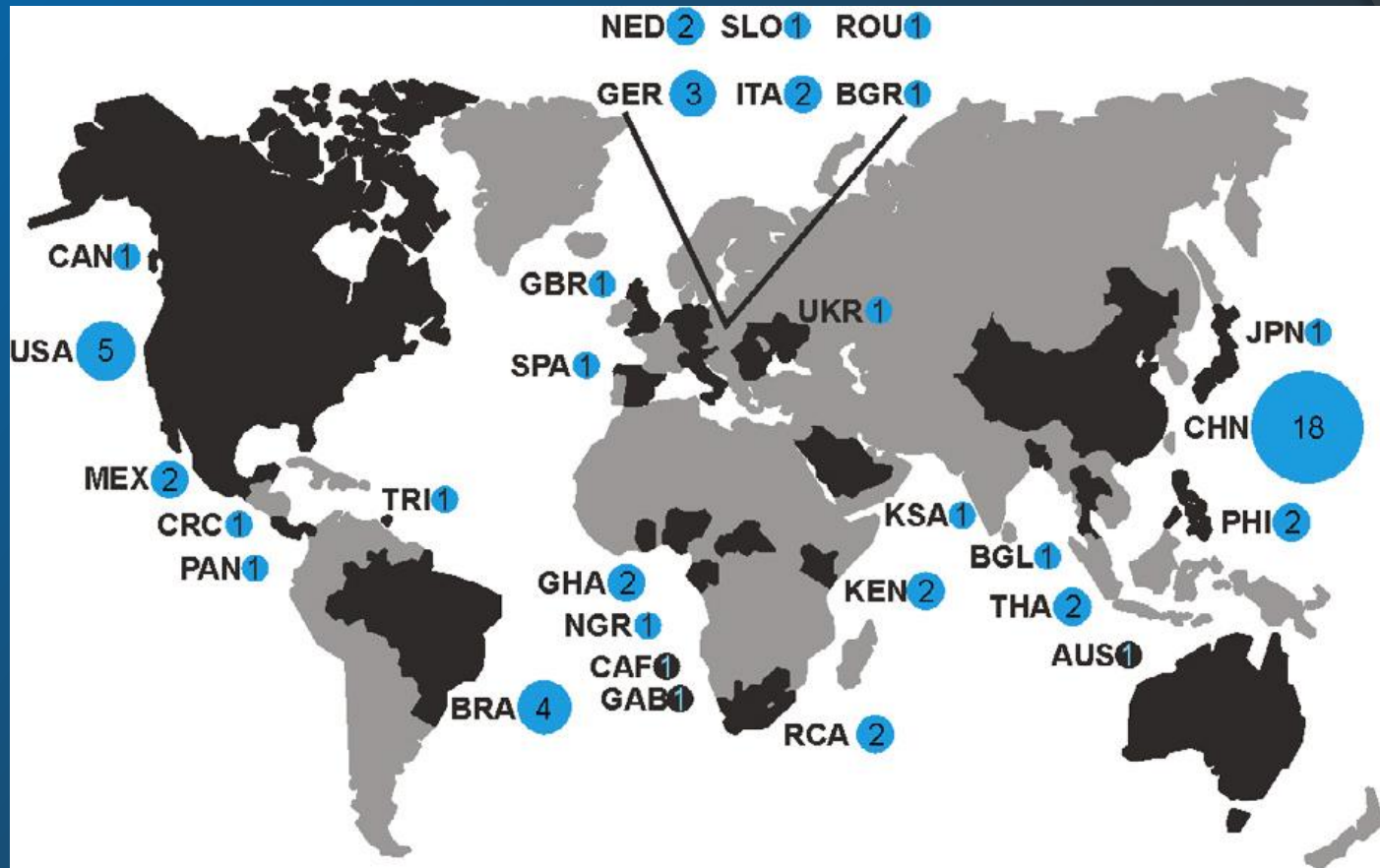
SEARCH FOR ZOONOTIC PATHOGENS ASSOCIATED WITH BATS AT THE HUMAN, DOMESTIC ANIMAL & WILDLIFE INTERFACES: SELECTED CASE STUDIES



WHY CONDUCT SUCH WORK?

- Poor surveillance in developing countries.
- A probability of importation of pathogens between continents.
- A possibility of exposure to tourists and workers travelling overseas.
- Lack of ideal immunological cross reactivity, provided by current biologics against certain agents.
- Recognition of ecological utility and imperiled status of bat communities on a global basis.

Coronaviruses in bats

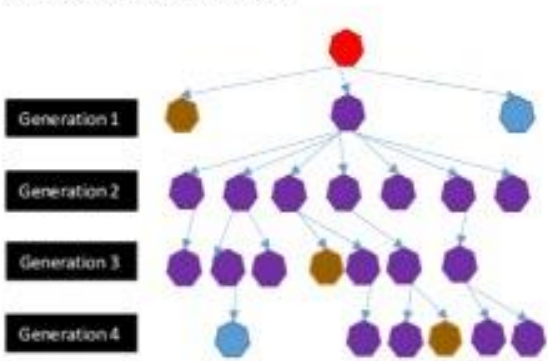


Bats are recognized as hosts of coronaviruses (CoV) on different continents. The biodiversity of CoVs suggests that bats may be the primary hosts where alpha- and betacoronaviruses evolved, including at least 3 of the 6 known human CoVs (Drexler et al., 2014).

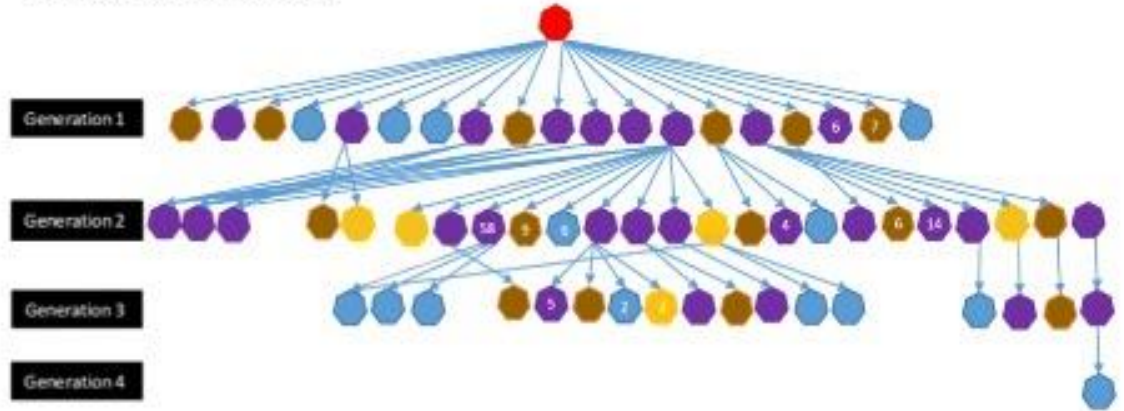
- Evidence points to the SARS CoV as originating from Chinese bats (presumably of the *Rhinolophus* genus) where anthropogenic factors promoted interspecies transmission to other mammals and thereafter to humans.
- Further studies performed in China and Hong Kong led to identification of several novel CoV groups in various bat species (Woo *et al.*, 2006).
- Specific drivers and mechanisms of CoV host shifts and switches, as well as associated evolutionary changes, are unknown.



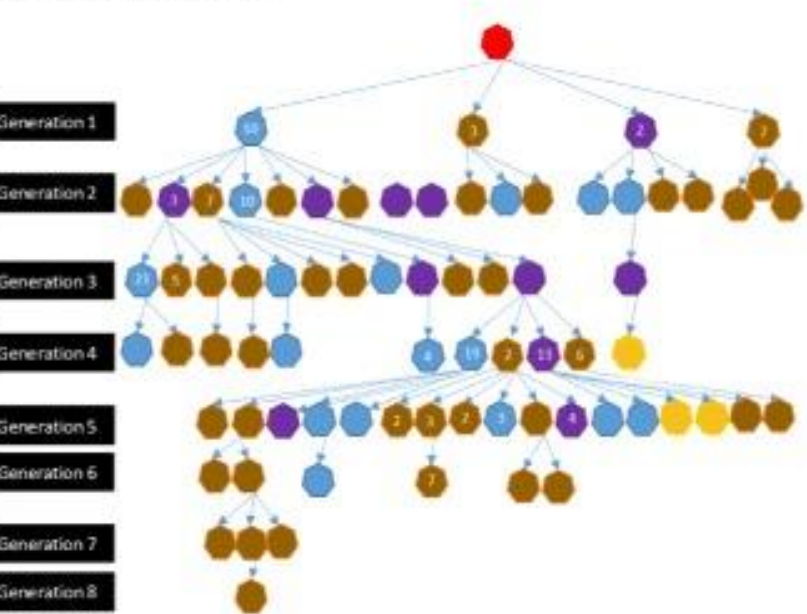
A MERS-CoV: Al-Hasa



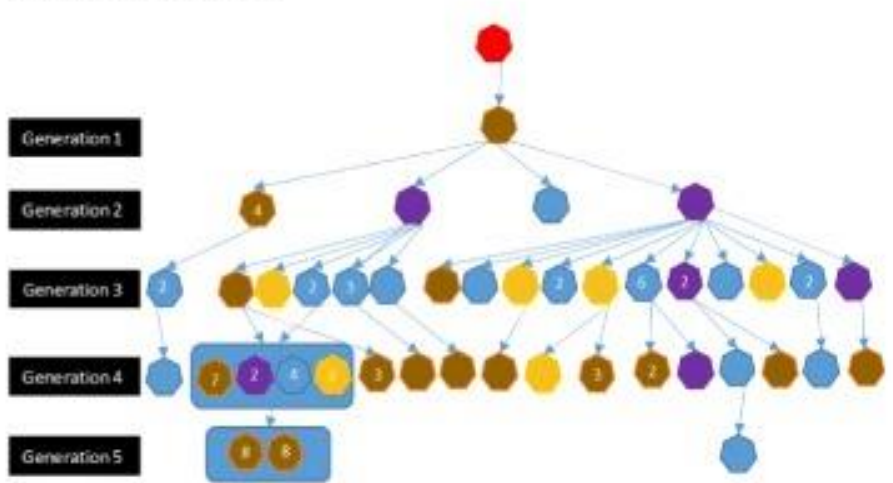
B MERS-CoV: South Korea



C SARS-CoV: Singapore



D SARS-CoV: Toronto



 Index Case

 Healthcare Worker

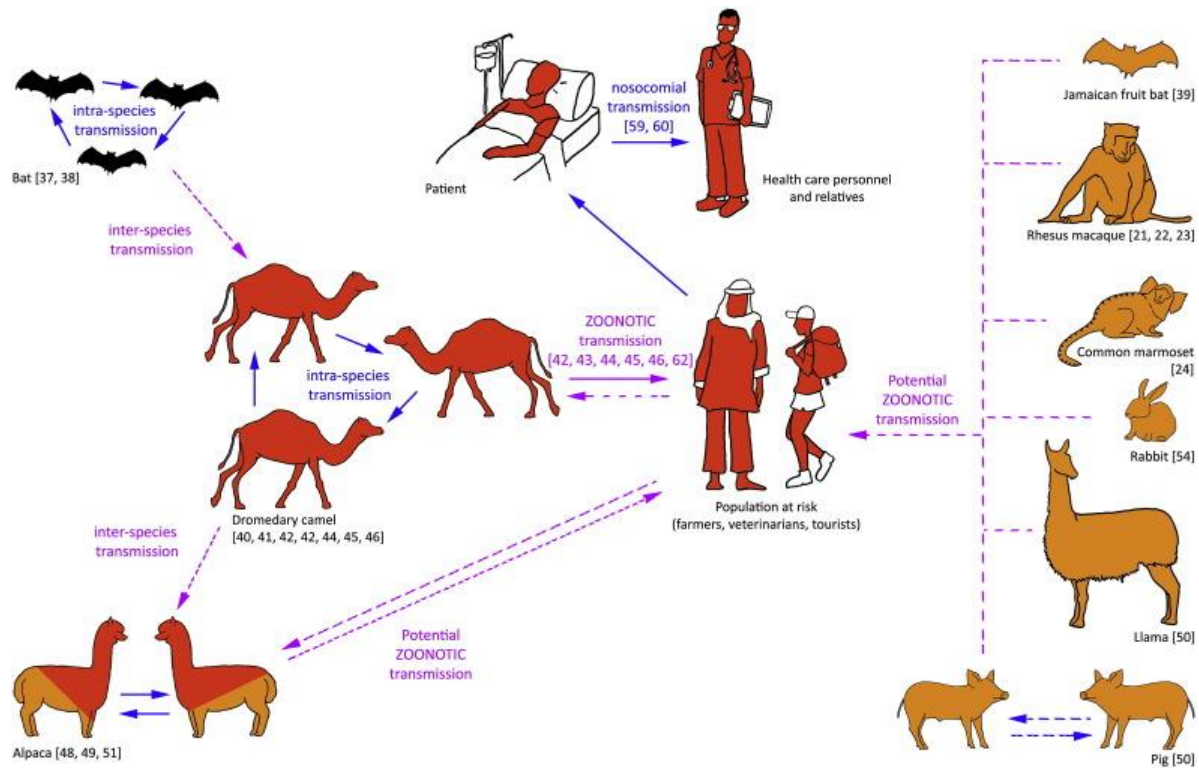
 Patient

 Family or Visitor

 Other-Healthcare Worker

 Cluster of cases shared by two cases from different generations

Middle Eastern Respiratory Syndrome (MERS) & Bats



Vergara-Alert, et al. One Health. 2017 Jun;3:34-40.

CASE REPORT, IN - DIAGNOSIS

- History of recent travel from Saudi Arabia
- Hospital worker
- Sputum, NP and oral swabs submitted
- DOH coordinated collection and shipment of specimens to CDC
- MERS CoV was identified by RT-PCR
- Recovered and discharged on ~ D12
- The man was the first US case with confirmed MERS CoV
- Contact tracing of hundreds of persons

Filoviruses in bats

To date, bats are the only major group, besides apes and humans, from which filoviruses were isolated, or viral RNA was detected (Uganda, Democratic Republic of the Congo, Gabon, Kenya, Spain).



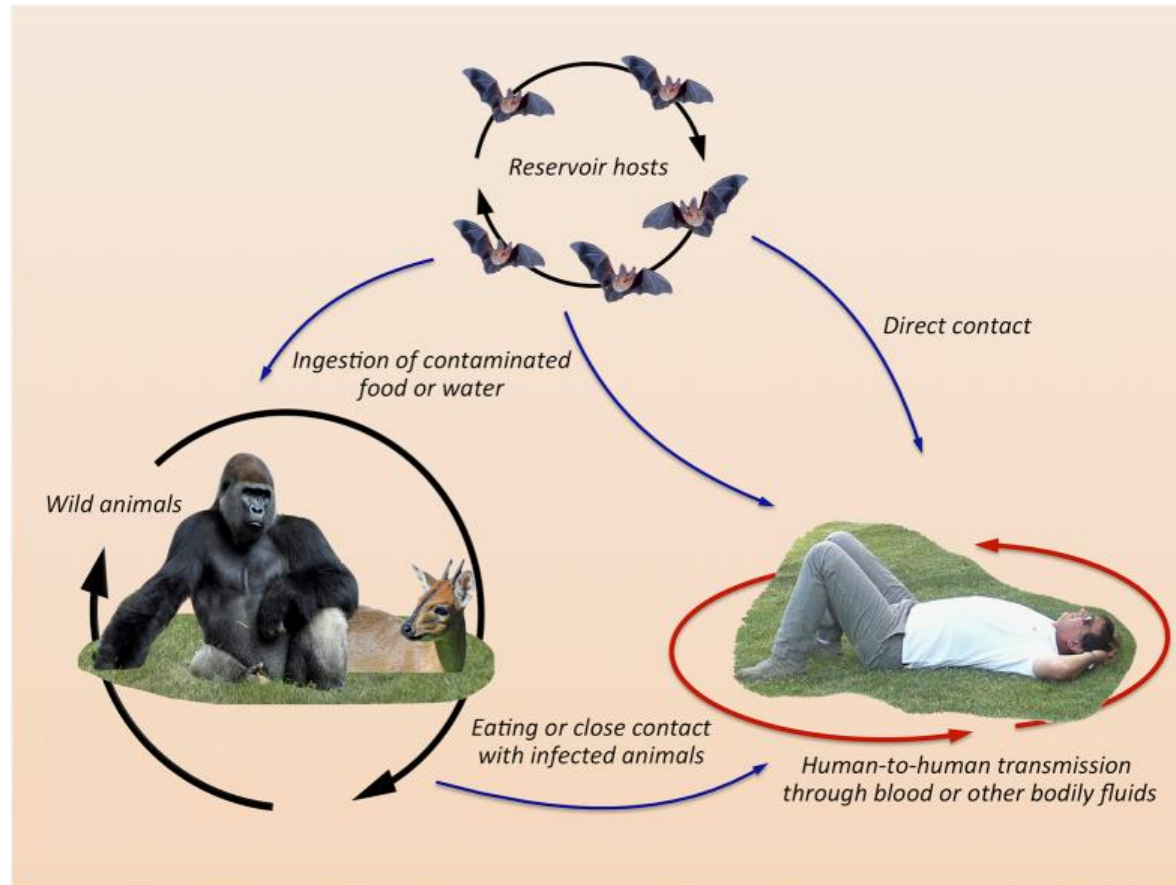
- EBOV RNA was detected in tree roosting fruit bats in Gabon: 19% *Hypsignathus monstrosus*; 4% *Epomops franqueti*; 3% *Myonycteris torquata*. Seroprevalence 24%, 7% and 7%, respectively (Leroy et al., 2005).
- An EBOV outbreak in DRC during 2007 was implicated as a result of direct exposure to migratory fruit bats (Leroy et al., 2009), however, the migratory fruit bat species such as *Eidolon helvum* were never diagnosed positive for EBOV.
- In Ghana, EBOV antibodies were detected in sera
Of 10/27 *Epomops franqueti*, 14/37 *Epomophorus gambianus*, 7/16 *Hypsignathus monstrosus*, and 1/4 *Nanonycteris veldkampii* fruit bats (Hayman et al., 2012).

- In Uganda, 5.1 % of *Rousettus aegyptiacus* fruit bats were positive for MARV RNA.
- From bats with a high RNA load, MARV has been isolated (liver).
- Seroprevalence of *R. aegyptiacus* to MARV was 2.4-9%.
- In addition, MARV RNA detected in 1/609 (0.2%) insectivorous bats *Hipposideros* sp (Towner et al., 2007; 2009).
- In another study, MARV RNA found in 3.0-3.6% of two species of insectivorous bats (*Rhinolophus eloquens* and *Miniopterus inflatus*) and in *R. aegyptiacus*, seroprevalence up to 20% (Swanepoel et al., 2007).



I. Kuzmin

WILDLIFE, BUSHMEAT & HUMAN TRANSMISSION



Allocati, et al. Cell Death Discov. 2016;2:16048.

CASE REPORT, CO - DIAGNOSIS

- 6 months post-presentation requested repeat evaluation and testing
- Patient learned of death in Dutch tourist from Marburg, that visited same bat cave in Uganda
- Serum tested positive for anti-MARV IgG by ELISA (1st filo-VHF in the USA), prompting additional testing of her archived D10 serum
- Traditional reverse-transcriptase polymerase chain reaction (RT-PCR) was negative; real-time (Taqman) RT-PCR equivocal; nested RT-PCR confirmed presence of MARV RNA fragments in D 10 sample

Bat paramyxoviruses

Henipaviruses

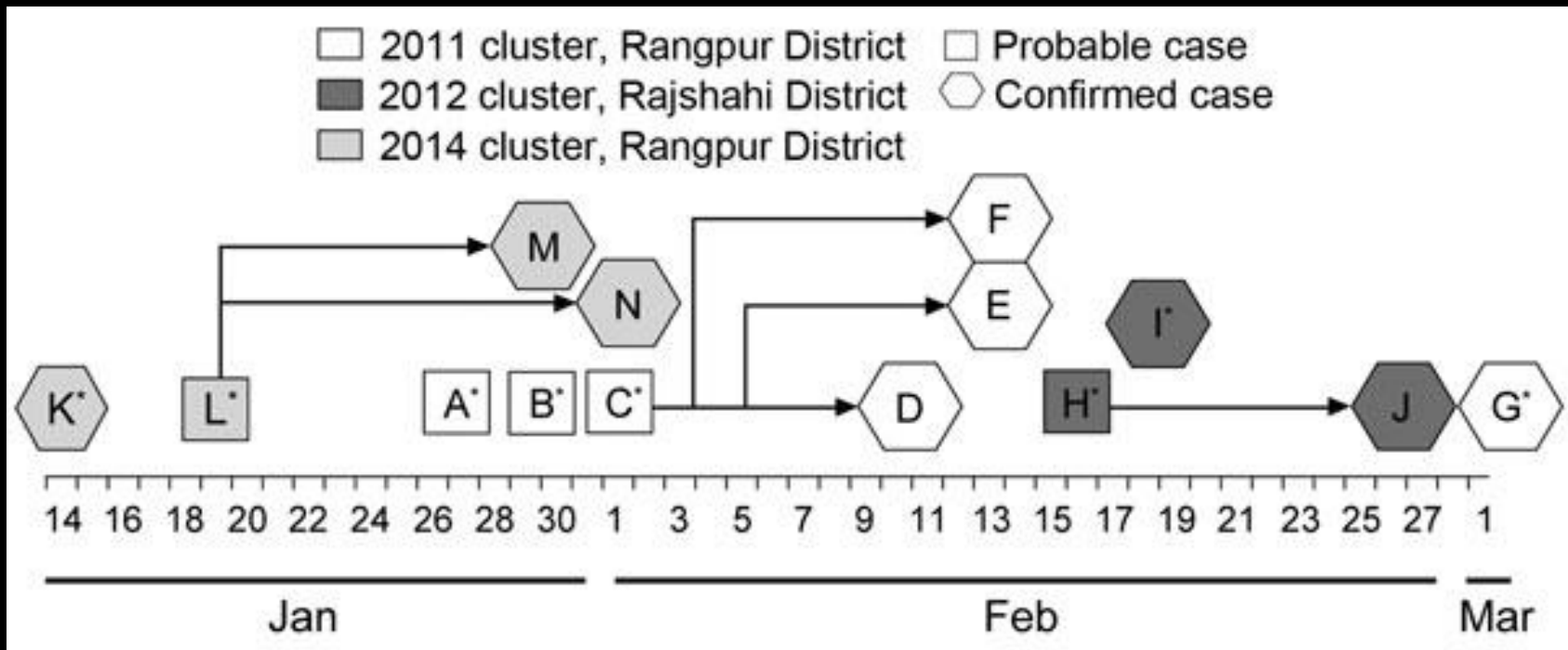
- Hendra virus (HeV) was identified in Australia in 1994. Fruit bats (*Pteropus* spp) are the principal hosts, whereas horses were the amplifying hosts and source of HeV infection (encephalitis with high fatality rate) for humans (Westbury, 2000).
- Nipah virus (NiV) was identified in Malaysia in 1999. Fruit bats (*Pteropus* spp) are the principal hosts, and pigs were the amplifying hosts and source of NiV infection for humans, with >300 cases (Uppal, 2000).



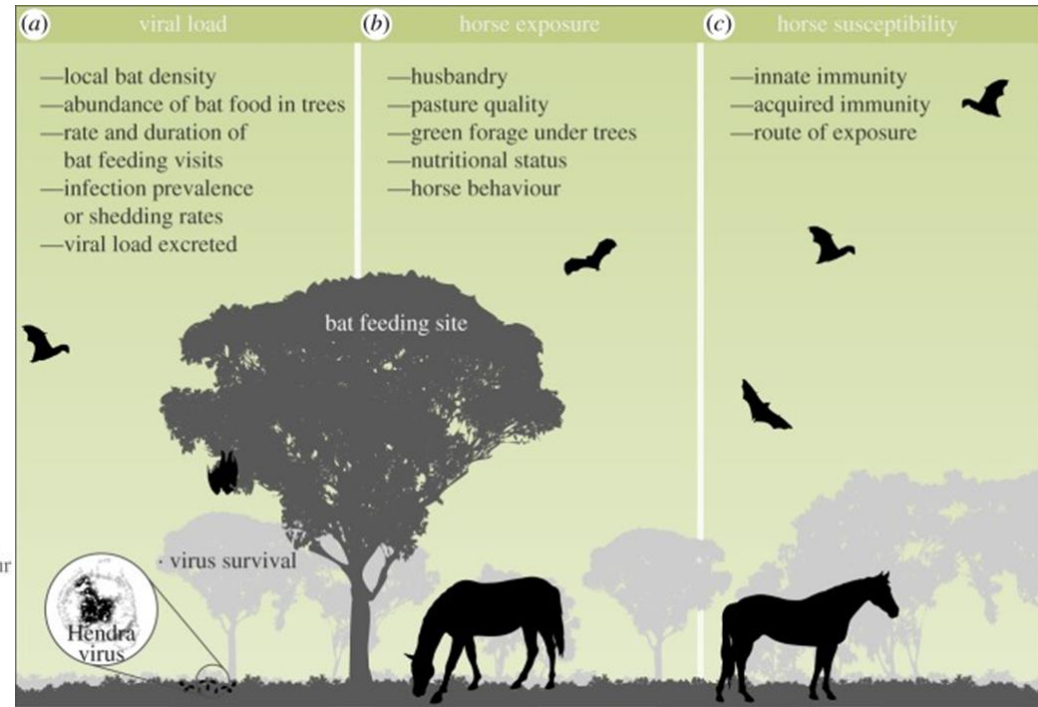
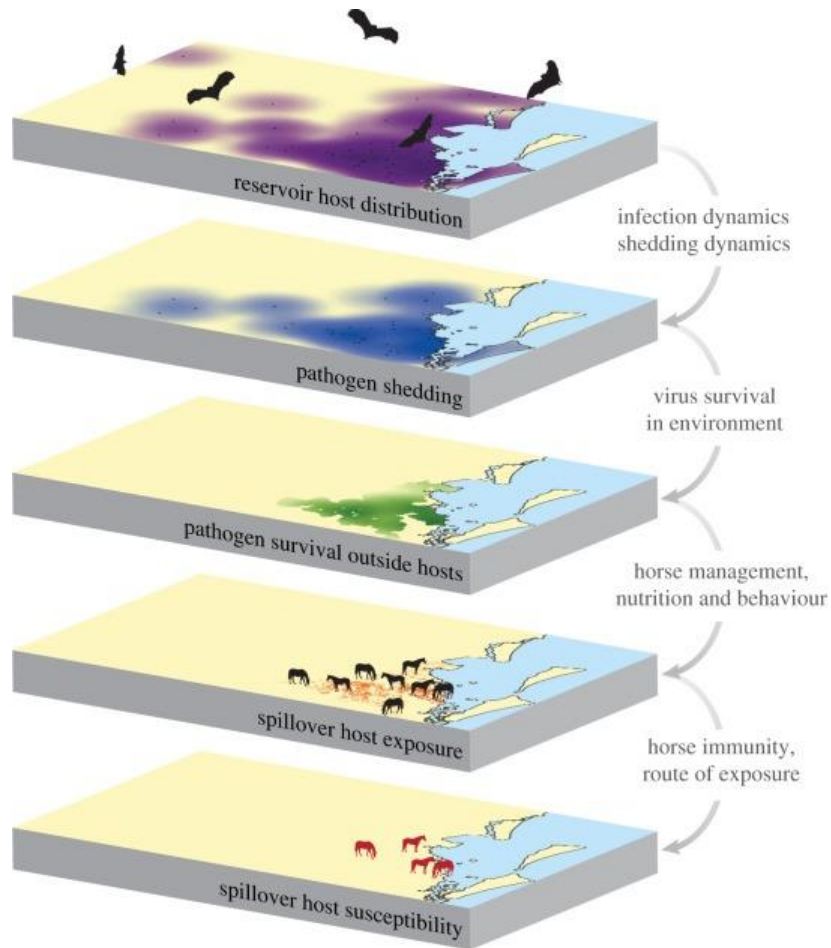
- In Bangladesh during 2001, NiV was transmitted to humans without apparent participation of an amplifying host, presumably directly from pteropid bats via raw date palm sap (*Montgomery et al., 2008*). Further, the virus can be transmitted human to human (*Sazzad et al., 2013*).
- Similar NiV outbreaks were documented in India (*Arankalle et al., 2011*).
- Antibodies to NiV were detected in *Pteropus* spp fruit bats in Bangladesh, Cambodia, India, Indonesia, Malaysia, and Thailand (seroprevalence 11-15%). In some of these cases NiV was isolated (*Johara, 2001; Olson et al 2002; Wacharapluesadee et al 2005; Epstein et al., 2008; Sendow et al 2009*).



A Timeline of Onset with Primary and Secondary Human Cases of Nipah Virus Infection -Bangladesh, 2011 - 2014



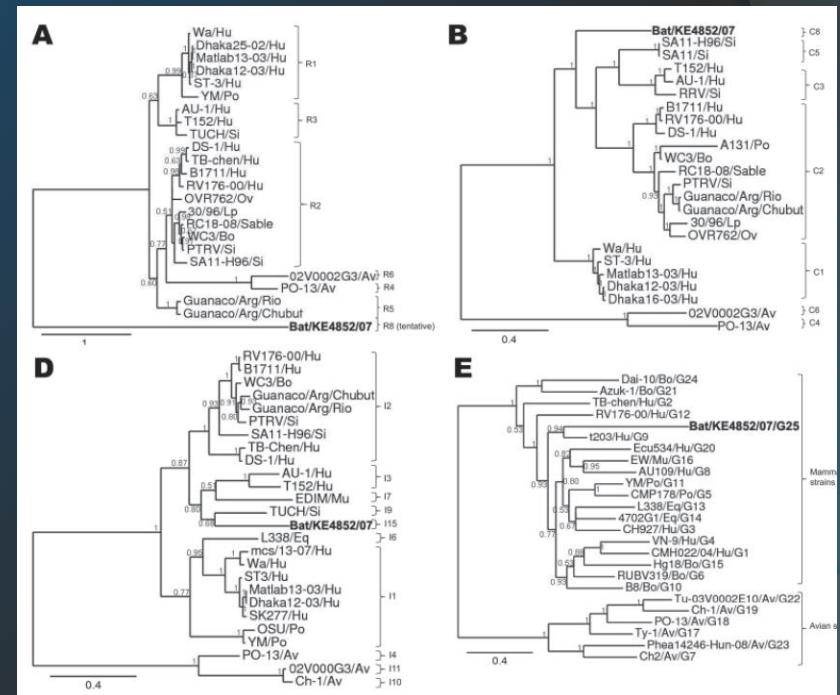
Plowright, et al. Proc Biol Sci. 2015 Jan 7;282(1798):20142124.



Ecological dynamics of emerging bat virus spillovers

A reassortant rotavirus

- A highly unusual rotavirus, with evidence of gene reassortment with human rotavirus, was detected in a fecal swab from a straw-colored fruit bat, *E. helvum*, from Kenya (*Esona et al., 2010*).
- If true, this is the first documented case of pathogen flow from humans to bats.
- It was speculated that bat exposure might occur via drinking of water from contaminated sources.



Bats & Many Other Agents...

Kohl C, et al. **Hervey virus**: Study on co-circulation with Henipaviruses in Pteropid bats within their distribution range from Australia to Africa. PLoS One. 2018 Feb 1;13(2):e0191933.

Mendenhall IH, et al. Influence of age and body condition on **astrovirus** infection of bats in Singapore: An evolutionary and epidemiological analysis. One Health. 2017 Oct 6;4:27-33.

Yadav P, et al. Isolation of **Tioman virus** from Pteropus giganteus bat in North-East region of India. Infect Genet Evol. 2016 Nov;45:224-229.

Mortlock M, et al. Novel **paramyxoviruses** in bats from Sub-Saharan Africa, 2007-2012. Emerg Infect Dis. 2015;21:1840-3.

Conrardy C, et al. Molecular detection of **adenoviruses** in bats from Kenya. Am J Trop Med Hyg. 2014;91:258-66.

Gu SH, et al. Molecular phylogeny of **hantaviruses** harbored by insectivorous bats in Côte d'Ivoire and Vietnam. Viruses. 2014;6:1897-910.

Tong S, et al. New world bats harbor diverse **influenza A** viruses. PLoS Pathog. 2013;9:e1003657.

Kading RC, et al. Isolation and molecular characterization of Fikirini **rhabdovirus**, a novel virus from a Kenyan bat. J Gen Virol. 2013;94:2393-8.

Quan PL, et al. Bats are a major natural reservoir for **hepaciviruses** and **pegiviruses**. Proc Natl Acad Sci U S A. 2013;110:8194-9.

Tao Y, et al. Discovery of diverse **polyomaviruses** in bats and the evolutionary history of the Polyomaviridae. J Gen Virol. 2013;94:738-48.

Kosoy M, et al. **Bartonella** spp. in bats, Kenya. Emerg Infect Dis. 2010;16:1875-81.

Emerging Infectious Diseases & *Special* Hosts

Bats are unique in their propensity to *host more zoonotic viruses* per species, based on a comparison with rodents.

Specific *life-history and ecological factors* promote zoonotic viral richness.

More zoonotic viruses are hosted by bat species whose distributions overlap with a greater number of other species in the same taxonomic order (*sympatry*).

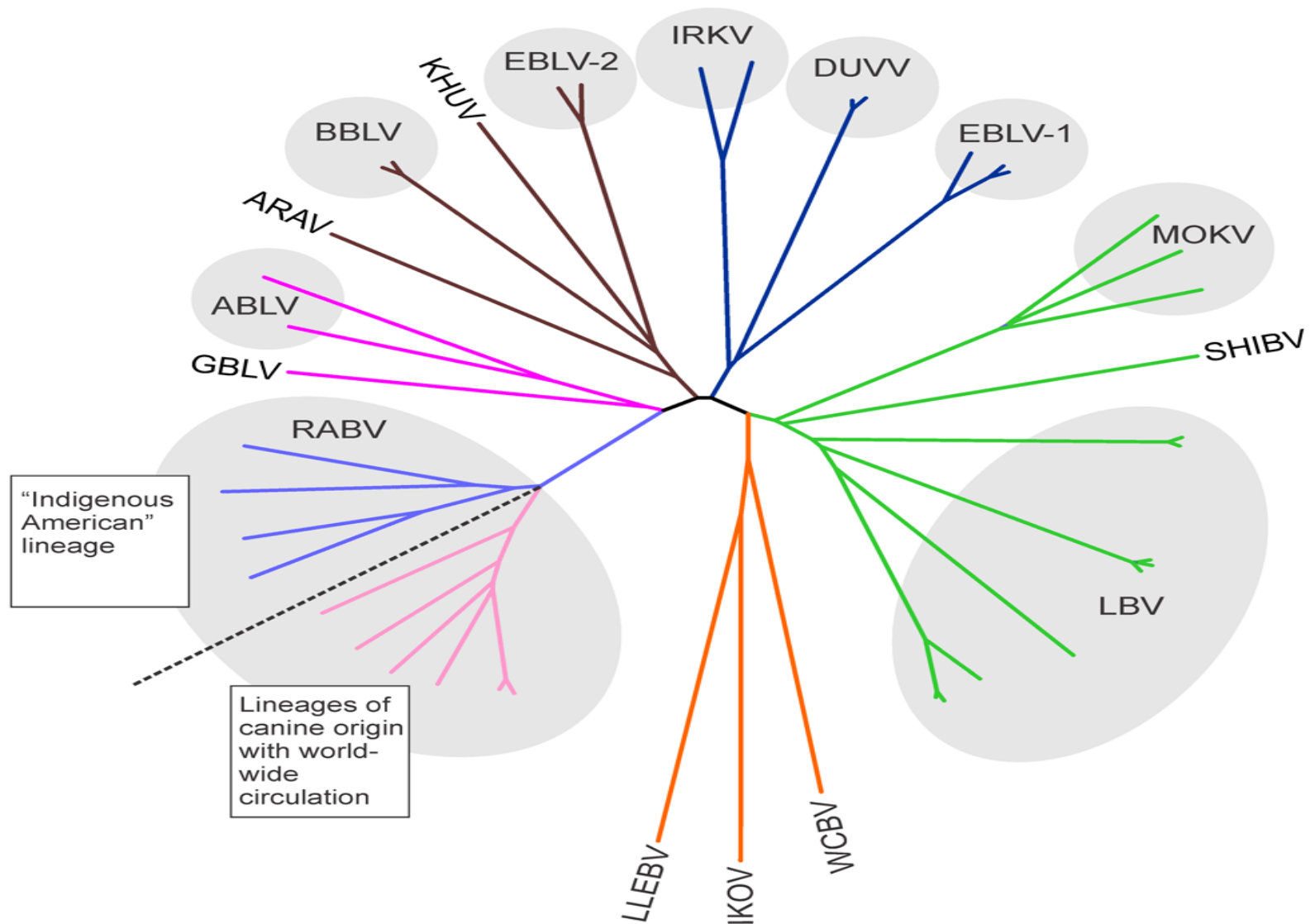
Evidence suggests *increased zoonotic viral richness* in species with *smaller litters* (one young), *greater longevity*, and *more litters* per year.

Inter-specific transmission is more prevalent among bats than among rodents.

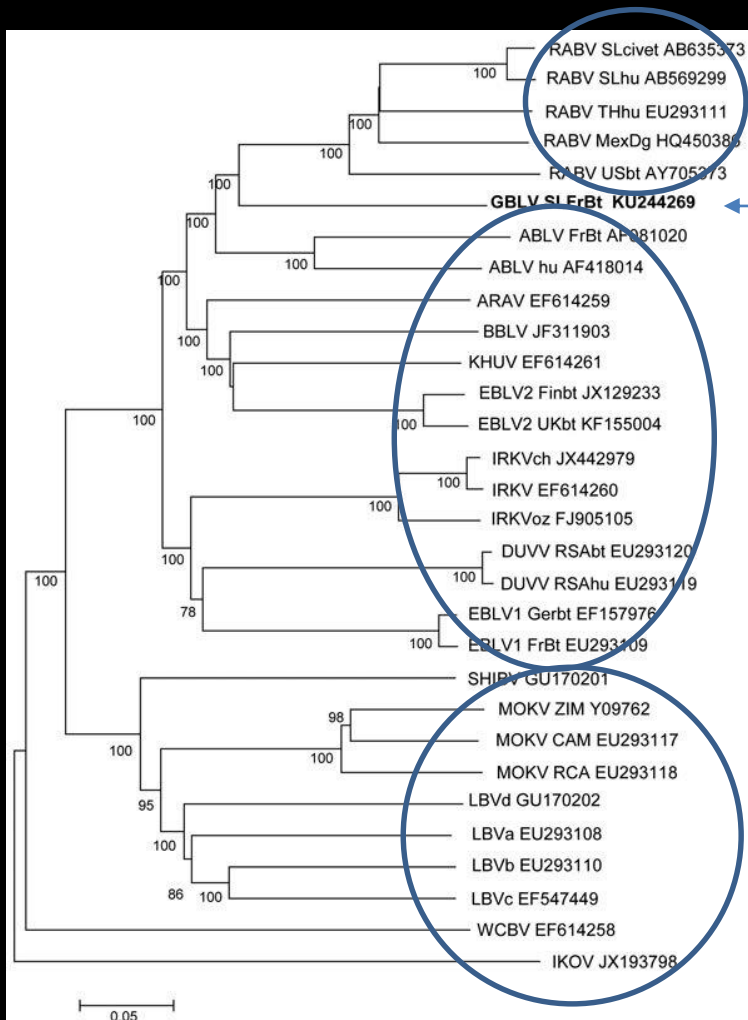
Findings shed light on disease emergence and perpetuation mechanisms and may lead to a predictive framework for identifying future reservoirs and outbreaks

Luis AD, et al. 2013 A comparison of bats and rodents as reservoirs of zoonotic viruses: are

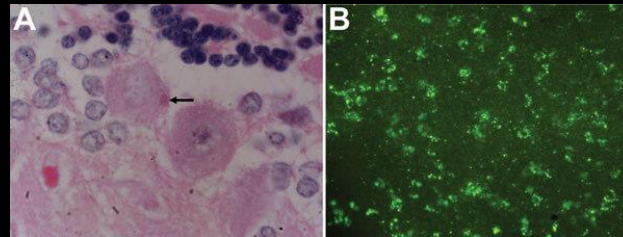
Extant Lyssavirus Phylogeny



A RATHER COMPLICATED PHYLOGENY...



Phylogenetic relationships between representatives from all classified lyssaviruses and novel **Gannoruwa** bat lyssavirus (GBLV) on the basis of complete genome sequences. Gunawardena et al. Emerg Infect Dis. 2016 Aug; 22(8): 1456–1459.



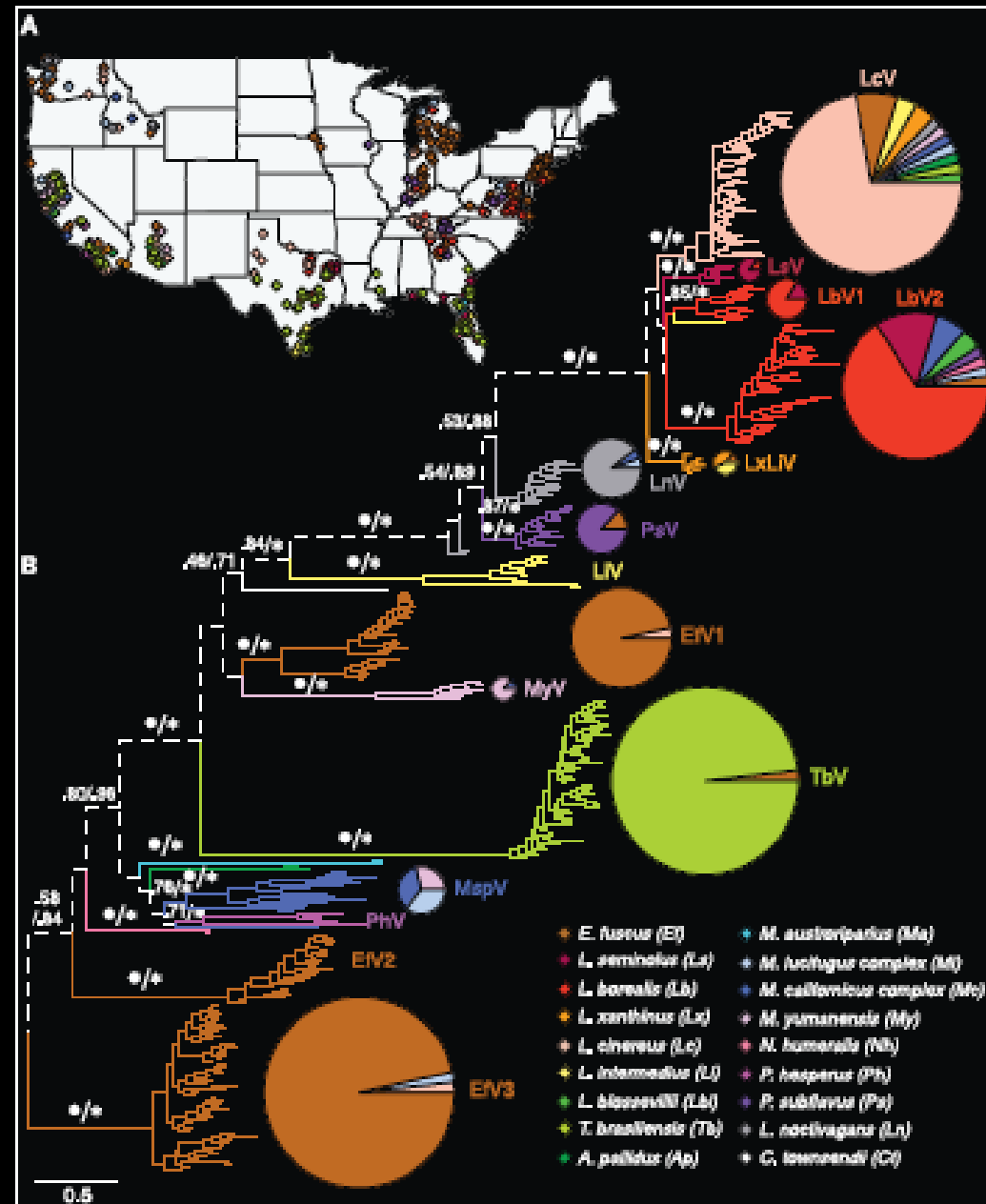
Negri bodies and lyssavirus antigens in brain tissue from an Indian flying fox, Sri Lanka

Bats and Rabies

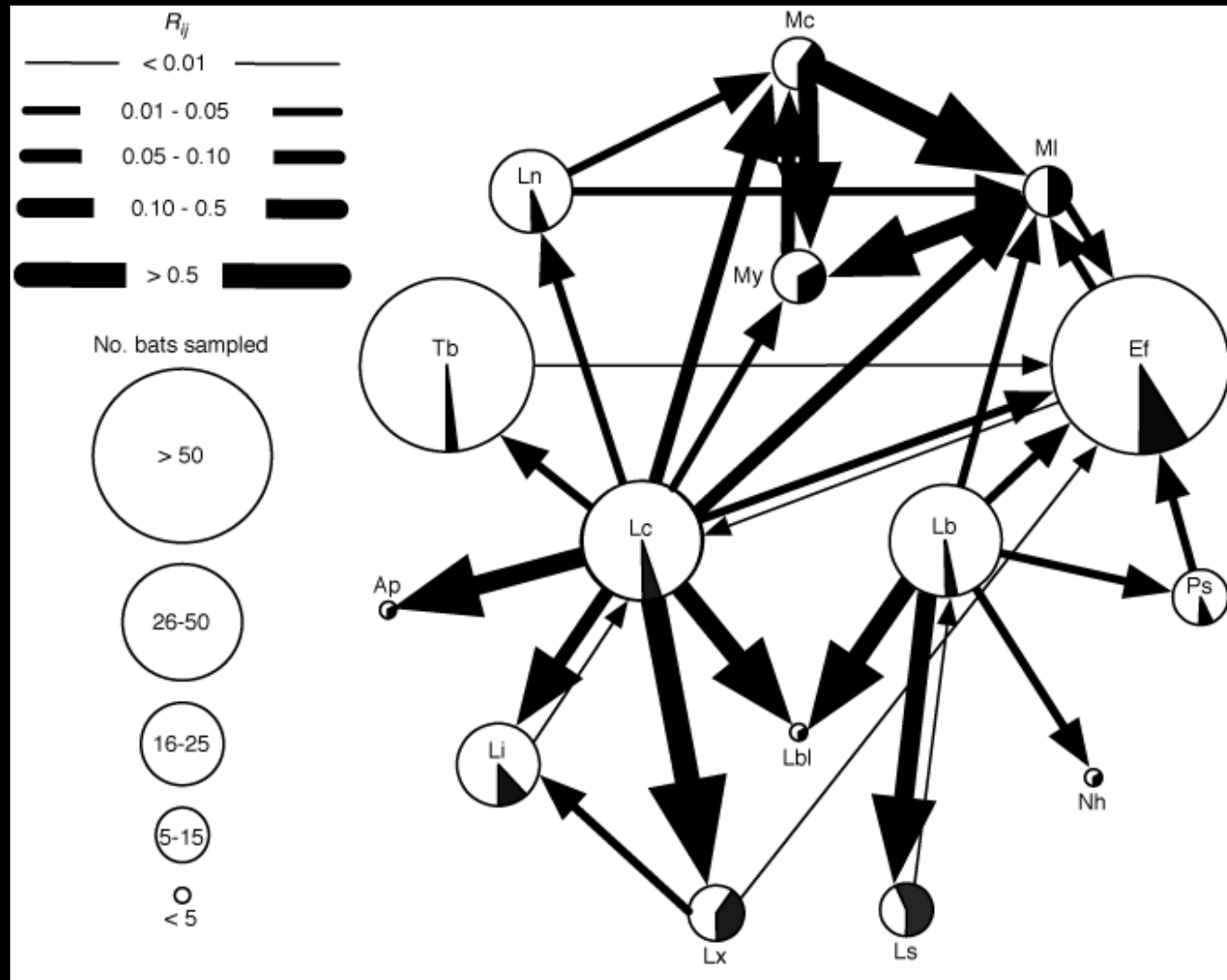
In the New World, only one lyssavirus, rabies virus, has been detected to date.

Among bats, evidence of many species-specific variants perpetuated in given taxa, but varying levels of inter-specific spillover events

Streicker et al. Science 2010



Community Ecology of Bat Pathogens

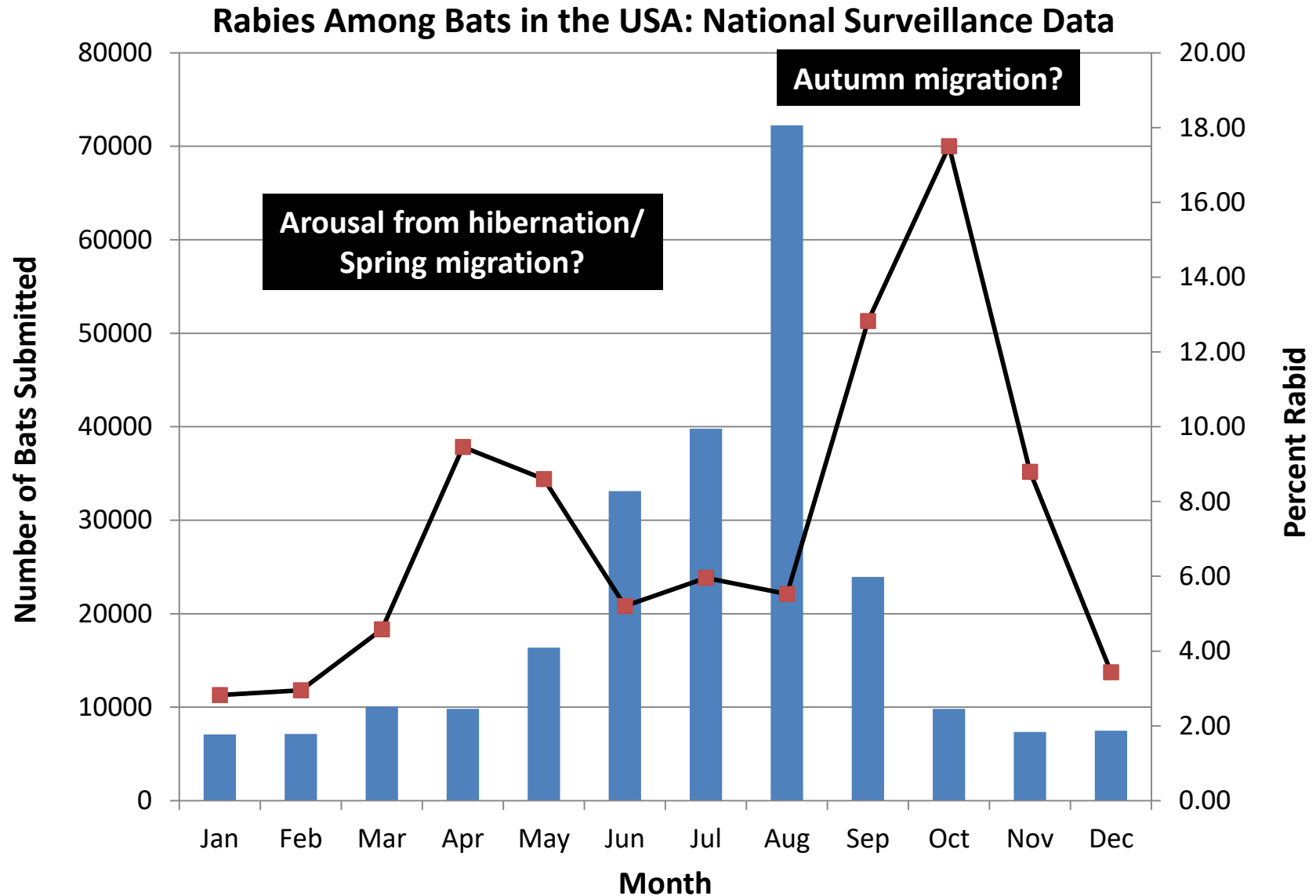


Bats and Rabies in the USA

- ❑ Between 2001 – 2009, >205,000 bats were submitted for rabies virus diagnosis.
- ❑ More than 13,000 (~7%) were found rabid.
- ❑ Bats represented ~22% of the 63,356 rabid animals diagnosed.
- ❑ All states, with the exception of Hawaii, were represented, but the Southwest had the highest proportionate rabies burden.
- ❑ Samples included > 40 native species, but >85% were represented by *Eptesicus* or *Myotis* taxa.
- ❑ Bats with the highest % rabid proportion included *Tadarida*, *Lasiurus*, and *Parastrellus*.
- ❑ Most bats (~59%) were submitted during summer, but the majority were found rabid during *autumn* (Odds Ratio 5.5)
- ❑ Bat species with *less conspicuous* roosting habits were significantly *more likely to be found rabid* (Odds Ratio 13.2)

Patyk et al., 2012

Bat Biology Influences Disease Patterns



Questions:

- Where did the host shift/switch of bat rabies virus to carnivores occur?
- When did it happen?
- Why did it result in sustained transmission?
- How frequently may it be going on today?

Recent host shifts of bat rabies viruses in North America

Kuzmin IV et al. PLoS
Pathog. 2012;8(6):e1002786.

AZ, 2001-2009

Outbreaks in striped skunks in Flagstaff area (2001; 2004-2005), and in gray foxes (2008-2009), caused by the big brown bat RABV.

USA, Mexico: 2008

Outbreak in white-nosed coatis in Cancun, Mexico. The same virus variant was detected in an immigrant in CA, who was exposed to a coati or fox in Oaxaca, Mexico. The virus was close but not identical to the *Tadarida brasiliensis* variant.

OR, 2009-2010

Two gray foxes and a goat at the end of winter, were infected with the same RABV variants, associated with *Myotis* bats and with big brown bats.

Human Rabies in the USA

- In ~ 21 of 36 cases where a bat virus variant was identified, it was the variant associated with silver-haired (*Lasionycteris noctivagans*) and eastern tri-colored (*Perimyotis subflavus*) bats.
- The rabies virus associated with Mexican free-tailed bats (*Tadarida brasiliensis*) was identified in ~10 cases.
- The viruses associated with big brown bat (*Eptesicus fuscus*) and *Myotis* spp bats caused human rabies in at least one case each.

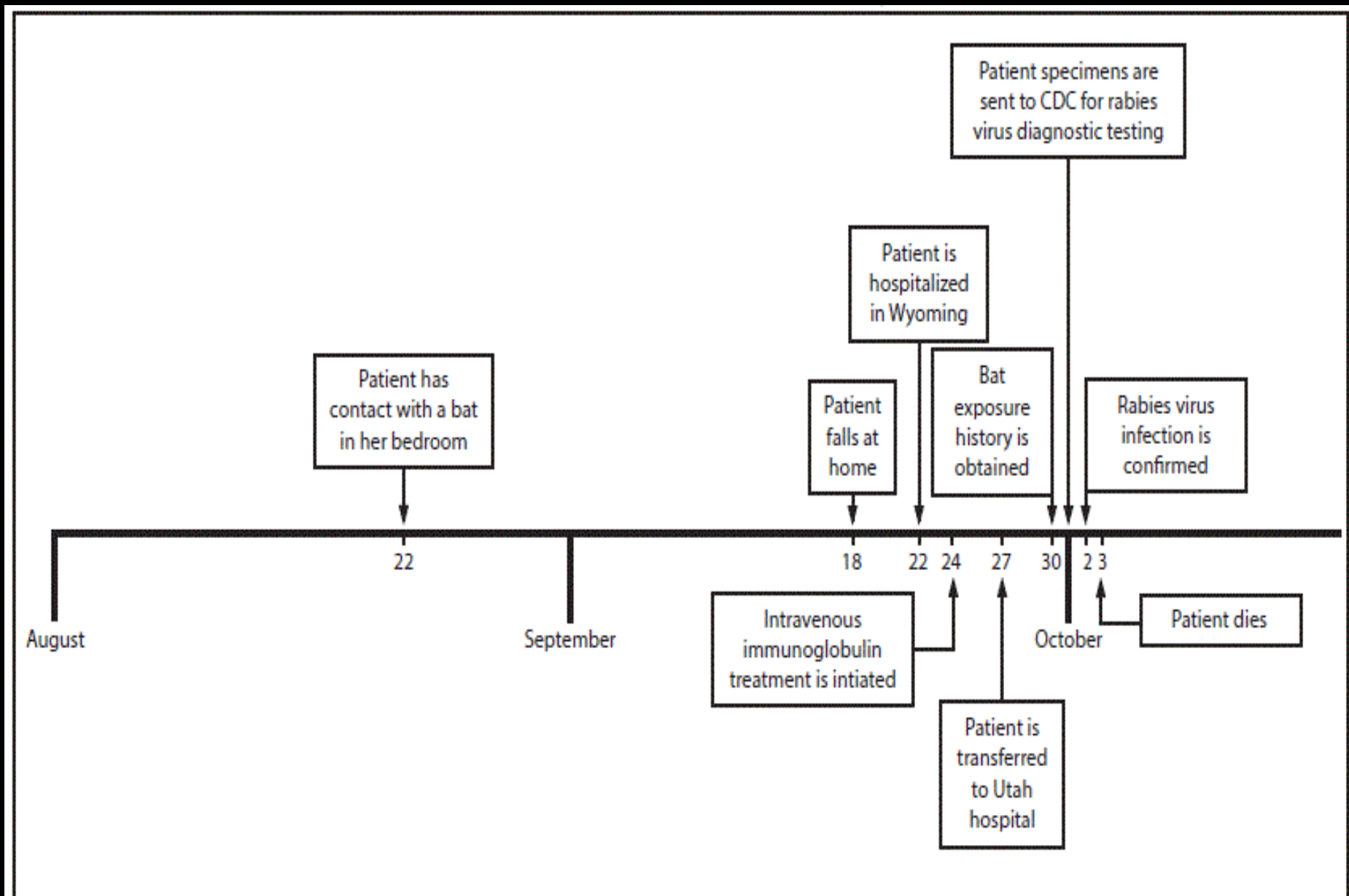
Greater susceptibility?

Ability of the virus to replicate in fibroblasts and epithelial cells?
(Morimoto et al., 1996)

CASE REPORT, WY - DIAGNOSIS

- D8 (retrospective): family members recalled that during August, the patient awoke at night in her home with a bat on her neck, which she swatted away with her hand and immediately washed her hand with soap and water
- Husband examined her for bite wounds and found none
- She did not seek medical attention
- Husband captured the bat with gloved hands and released it outside
- DOH coordinated collection and shipment of specimens to CDC
- Rabies virus RNA was detected in nuchal skin biopsy and saliva by RT/PCR
- Rabies virus antigens observed in nuchal skin biopsy by DFA test
- Rabies virus IgM and IgG detected in CSF and serum by IFA
- Rabies VNA confirmed by the RFFIT
- Rabies virus variant identified as enzootic to the silver-haired bat (*Lasionycteris noctivagans*).
- The woman was the first WY resident with confirmed rabies virus infection since the state began documenting reportable infectious diseases in 1911

TIMELINE (MMWR 2016, 65;529–533)

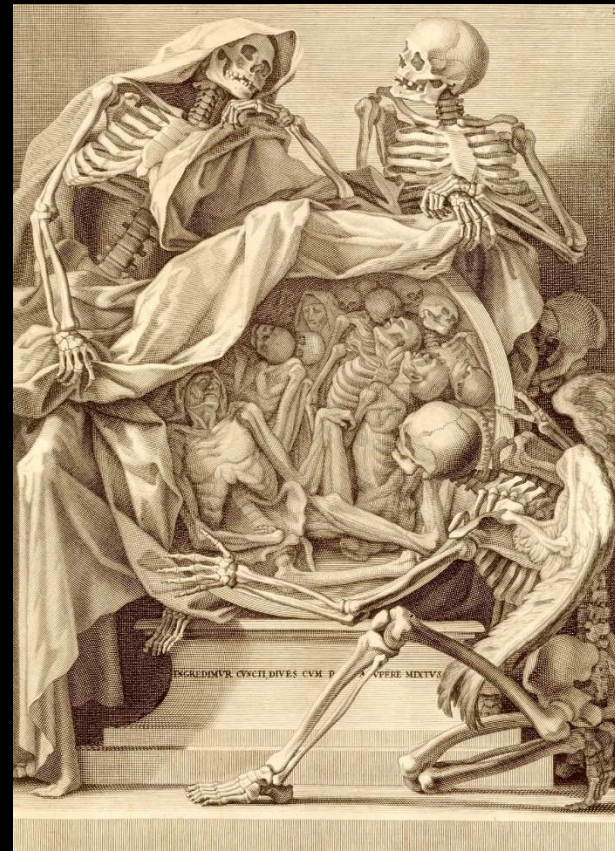


REPERCUSSIONS OF MIS-DIAGNOSIS: HUMAN TO HUMAN TRANSMISSION OF BAT RABIES VIRUS

Burton et al.

Arch Neurol. 2005 Jun;62(6):873-82.

Rabies encephalomyelitis: clinical, neuroradiological, and pathological findings in 4 transplant recipients.



BAT RABIES MANAGEMENT WITH POTENTIAL APPLICABILITY TO OTHER PATHOGENS?

Focused studies to understand transmission, perpetuation, and exacerbation

Public education concerning health risks and defining exposures

Outreach focusing upon ecological benefits, and detriments from population reduction

Training, PPE, and pre-exposure immunization for those at risk of exposure

Euthanasia of suspect animals and prophylaxis of exposed humans and other mammals

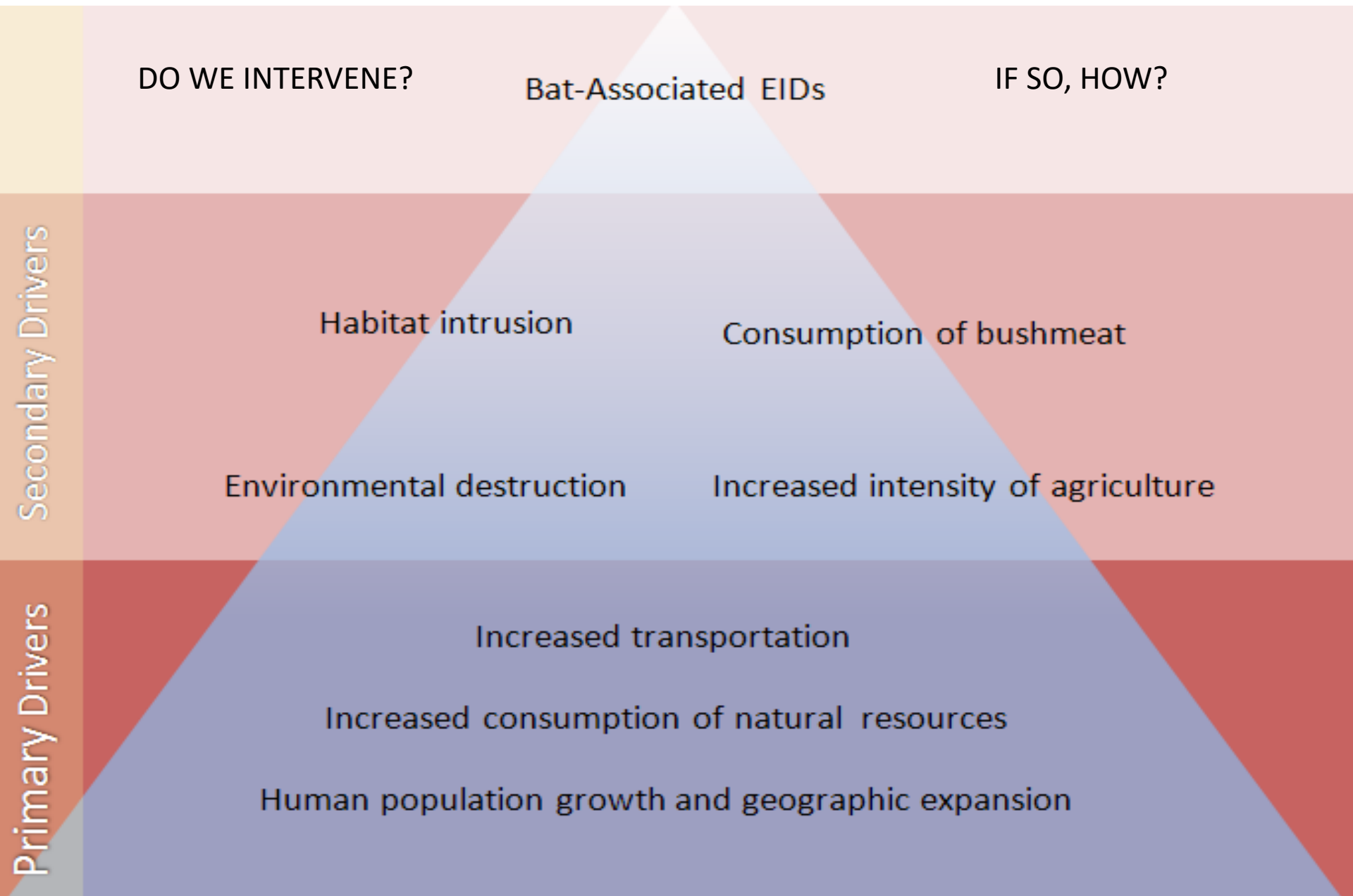
Community planning to mitigate impacts from inappropriate development

Exclusion of bats from human dwellings using appropriate methods

Syndrome-specific techniques to enhance local surveillance /control

Are We Approaching, At, Or Past A Critical Tipping Point in the *Anthropocene*?

Kuzmin IV et al. Bats, emerging infectious diseases, and the rabies paradigm revisited. Emerg Health Threats J. 2011 Jun 20;4:7159.



CONCLUSIONS

- Several newly appreciated emerging infectious diseases are associated with bats, due in part to their *distribution, abundance, diversity and vagility*.
- Bats DO appear '*special*' biologically, in their capacity to orchestrate various disease ensembles, compared to other mammals, such as rodents.
- Given their comparatively low reproductive potential, various '*stressors*' on extant bat communities may result in unpredictable ecological repercussions, due to their critical role in arthropod suppression, pollination, seed dispersal, and forest regeneration with unexpected consequences.
- *Rabies is the oldest known bat zoonosis* and multiple public health, agricultural, and wildlife biology '*lessons learned*' historically from this disease may be applicable directly to other emerging bat pathogens.
- A modern holistic *One Health strategy* integrating epidemiologically relevant best practices of disease prevention and control with the essential elements of conservation biology for wildlife management in general and for bats specifically should be espoused within a global context.

ACKNOWLEDGEMENTS

BAT CONSERVATION INTERNATIONAL, ESPECIALLY G. McCRACKEN

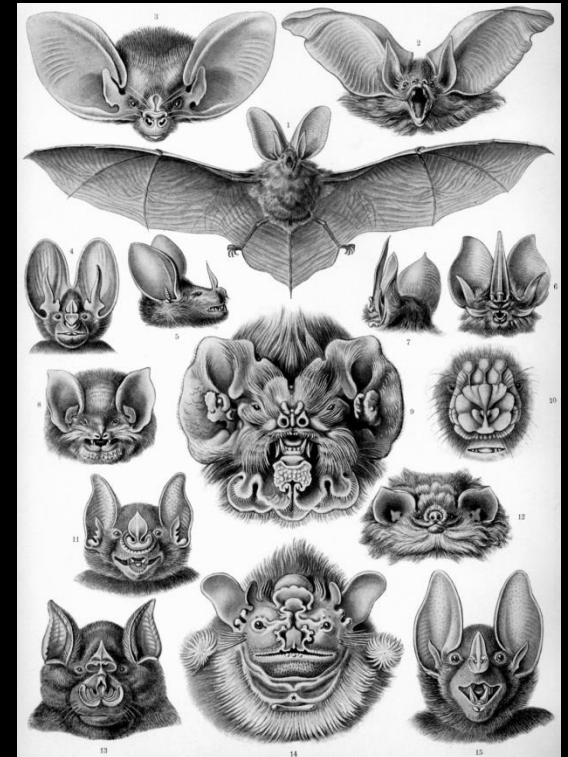
USGC, ESPECIALLY P. CRYAN, T. O'SHEA

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USDA, ESPECIALLY A. GILBERT

UNIVERSITY OF GLASGOW, ESPECIALLY D. STREICKER

CDC, OIE & WHO COLLEAGUES



QUESTIONS?

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