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Migrating Bats Are Basically Flying Weather Stations



George Dvorsky

9/20/17 3:55pm • Filed to: BATS ▾



11.0K



9



5



Common noctule (*Nyctalus noctula*). (Image: MPI f. Ornithology/ K. Safi)

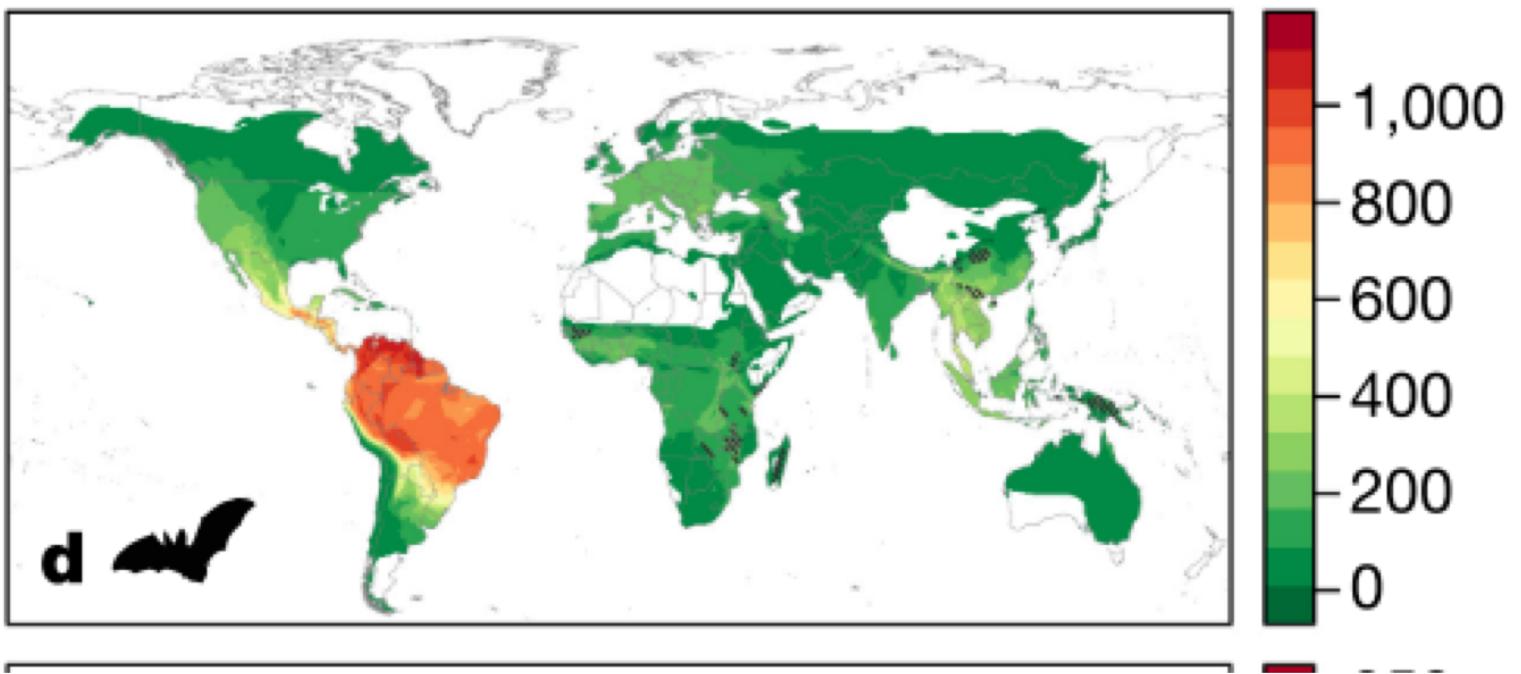
Bats

- ~1300 species (>10% of all mammals)
- Only mammal capable of flying
- Fastest active powered flight (McCracken 2016)
- Large diversity of foraging strategy
- Immune system



Zoonosis

- Most human emerging infectious diseases across the world are zoonotic
- Especially Viruses occurring in mammals are of major concern (i.e. Anthrax, Birdflu and Ebola (Karesh et al. 2012, Olival et al. 2018)
- Bats are carriers of a variety of zoonotic diseases such as filovirus, nile virus, ebola (Olival et al. 2017)



Global patterns of zoonosis

ARTICLE

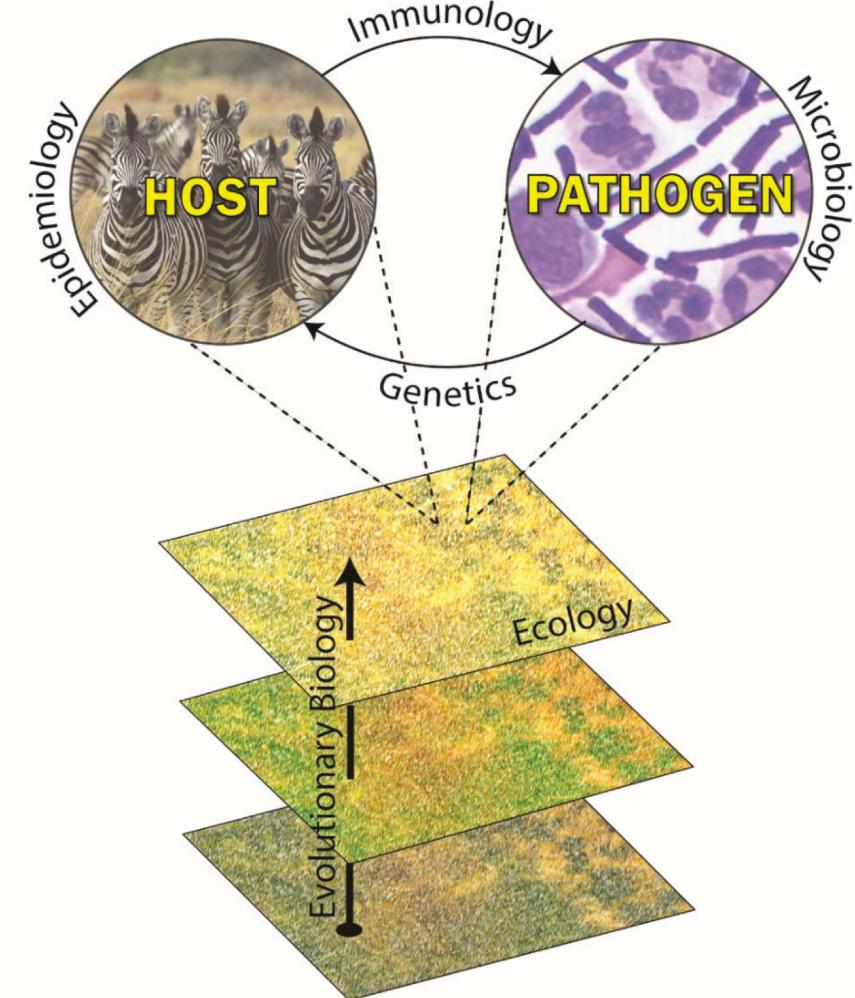
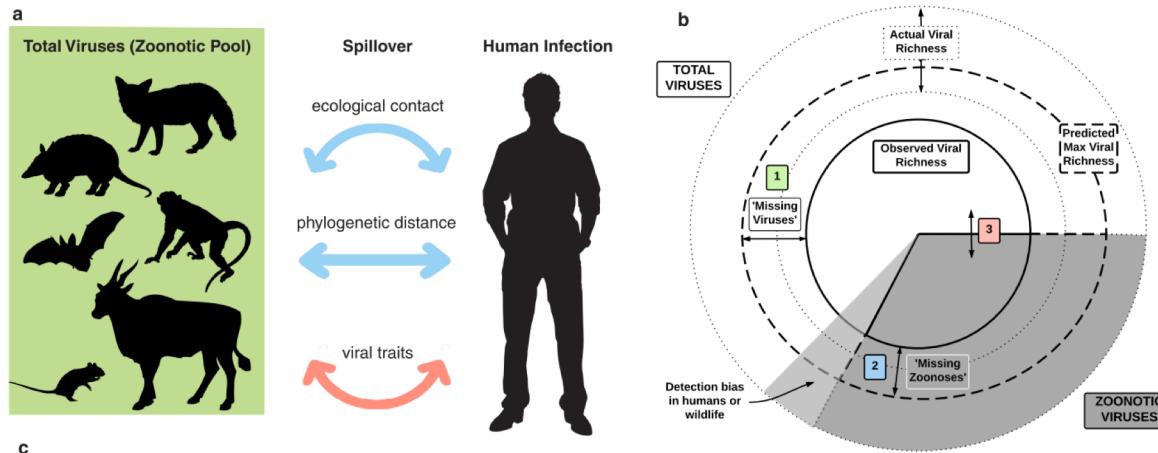
DOI: 10.1038/s41467-017-00923-8

OPEN

Global hotspots and correlates of emerging zoonotic diseases

Toph Allen¹, Kris A. Murray^{2,3}, Carlos Zambrana-Torrelio¹, Stephen S. Morse⁴, Carlo Rondinini⁵, Moreno Di Marco^{6,7}, Nathan Breit¹, Kevin J. Olival¹ & Peter Daszak¹

LETTER RESEARCH



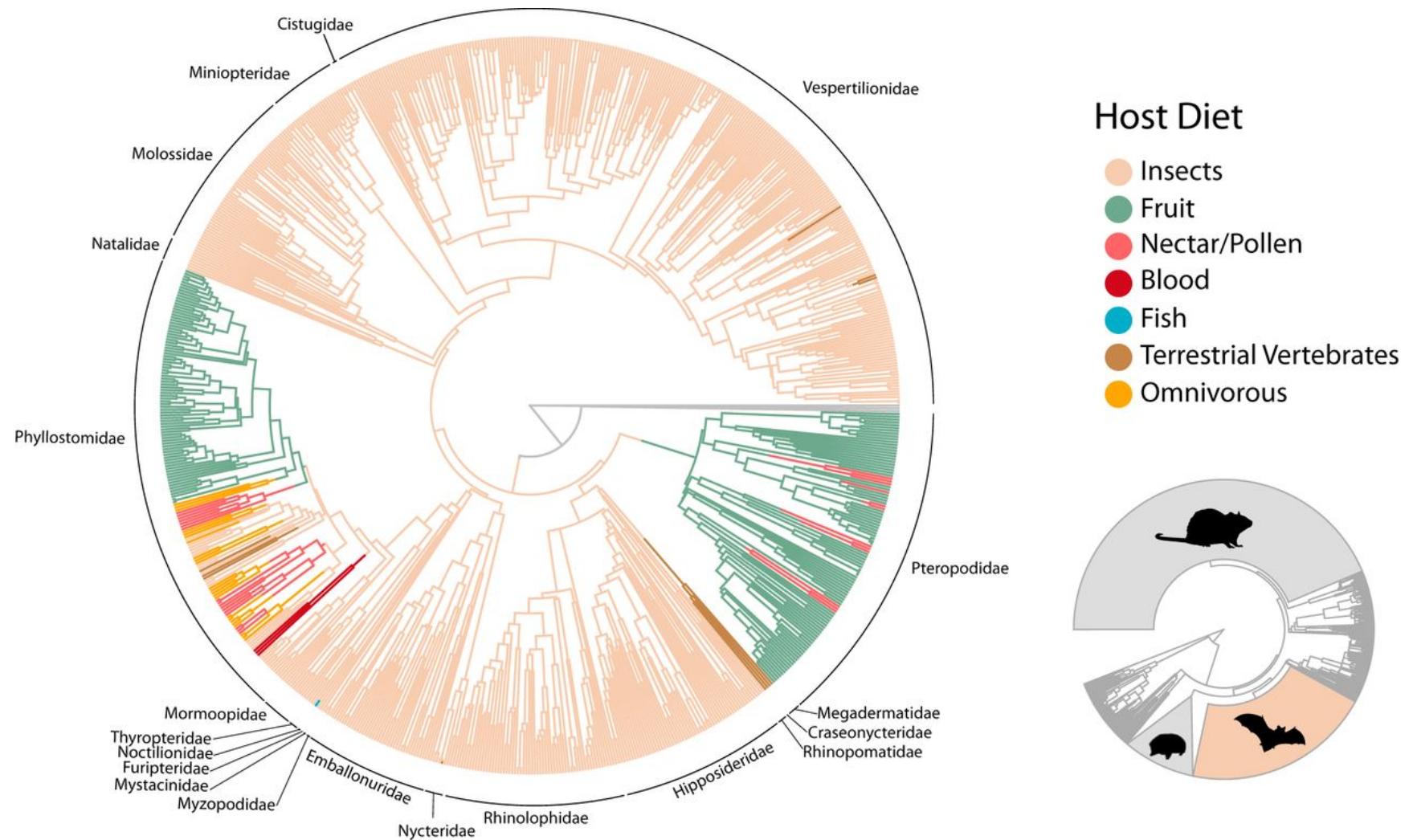
Carlson et al. 2017

Bats as zoonotic vectors

- It is important to understand how vectors of zoonosis have evolved their immune genes for difference against virulence, and key ecomorphological traits crucial for their survival

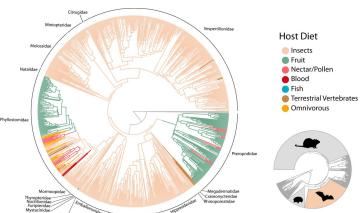
Motivation

The niche specialization of certain bat species might have been driven by the diversity of their innate immune genes, and key innovations- i.e diet, olfaction, echo location, flight (Hayden et al. 2014)



Motivation

The niche specialization of certain bat species might have been driven by the diversity of their innate immune genes, olfactory receptors (Hayden et al. 2014) and echo location genes (XXX).



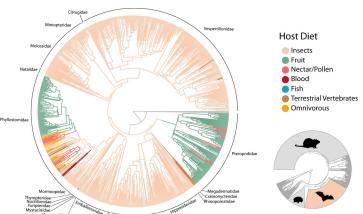
Annual Review of Animal Biosciences

Bat Biology, Genomes, and the Bat1K Project: To Generate Chromosome-Level Genomes for All Living Bat Species

Emma C. Teeling,¹ Sonja C. Vernes,^{2,3}
Liliana M. Dávalos,⁴ David A. Ray,⁵
M. Thomas P. Gilbert,^{6,7} Eugene Myers,⁸
and Bat1K Consortium*

Motivation

The niche specialization of certain bat species might have been driven by the diversity of their innate immune genes, olfactory receptors (Hayden et al. 2014) and echo location genes (XXX).



Annual Review of Animal Biosciences

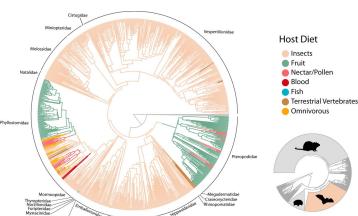
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Comparative Analysis of Bat Genomes Provides Insight into the Evolution of Flight and Immunity

Guojie Zhang,^{1,2*}† Christopher Cowled,^{3*} Zhengli Shi,^{4*} Zhiyong Huang,^{1*}
Kimberly A. Bishop-Lilly,^{5*} Xiaodong Fang,¹ James W. Wynne,³ Zhiqiang Xiong,¹
Michelle L. Baker,³ Wei Zhao,¹ Mary Tachedjian,³ Yabing Zhu,¹ Peng Zhou,^{3,4} Xuanting Jiang,¹
Justin Ng,³ Lan Yang,¹ Lijun Wu,⁴ Jin Xiao,¹ Yue Feng,¹ Yuanxin Chen,¹ Xiaoqing Sun,¹
Yong Zhang,¹ Glenn A. Marsh,³ Gary Crameri,³ Christopher C. Broder,⁶ Kenneth G. Frey,⁵
Lin-Fa Wang,^{3,7}† Jun Wang^{1,8,9}†

Toll like receptors in bats



Annual Review of Animal Biosciences

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Michelle L. Baker,⁸ Wei Zhao,⁹ Mary Tachedjian,³ Yabing Zhu,¹ Peng Zhou,^{3,10} Xuanting Jiang,¹
Justin Ng,² Lan Yang,² Lijun Wu,⁴ Jin Xiao,¹ Yu Feng,¹ Yuanxin Chen,¹ Xiaozheng Sun,¹
Yong Zhang,³ Glenn A. Marsh,³ Gary Cramer,³ Christopher C. Broder,⁶ Kenneth G. Frey,⁵
Lin-Fa Wang,^{3,7,†} Jun Wang,^{1,6,†}

Lineage	Symbol	Gene	ω_0 (average)	ω_1 (other)	ω_2 (target)	P value
Ancestor	TLR7	Toll-like receptor 7	0.2821	0.2670	2.7778	3.54E-07
	<i>ATM</i>	Ataxia telangiectasia mutated	0.20096	0.19595	0.7163	1.34E-05
	<i>MDM2</i>	Mdm2 p53 binding protein homolog (mouse)	0.13358	0.12615	0.81085	4.05E-04
	<i>NLRP3</i>	NLR family, pyrin domain-containing 3	0.1788	0.1714	1.1884	1.93E-04
	<i>MAP3K7</i>	Mitogen-activated protein kinase kinase kinase 7	0.0216	0.0194	0.4786	8.93E-03
	<i>RAD50</i>	RAD50 homolog	0.09657	0.09343	0.28882	7.95E-03
	<i>PRKDC</i>	Protein kinase, DNA-activated, catalytic polypeptide	0.23036	0.22768	0.45155	6.80E-03

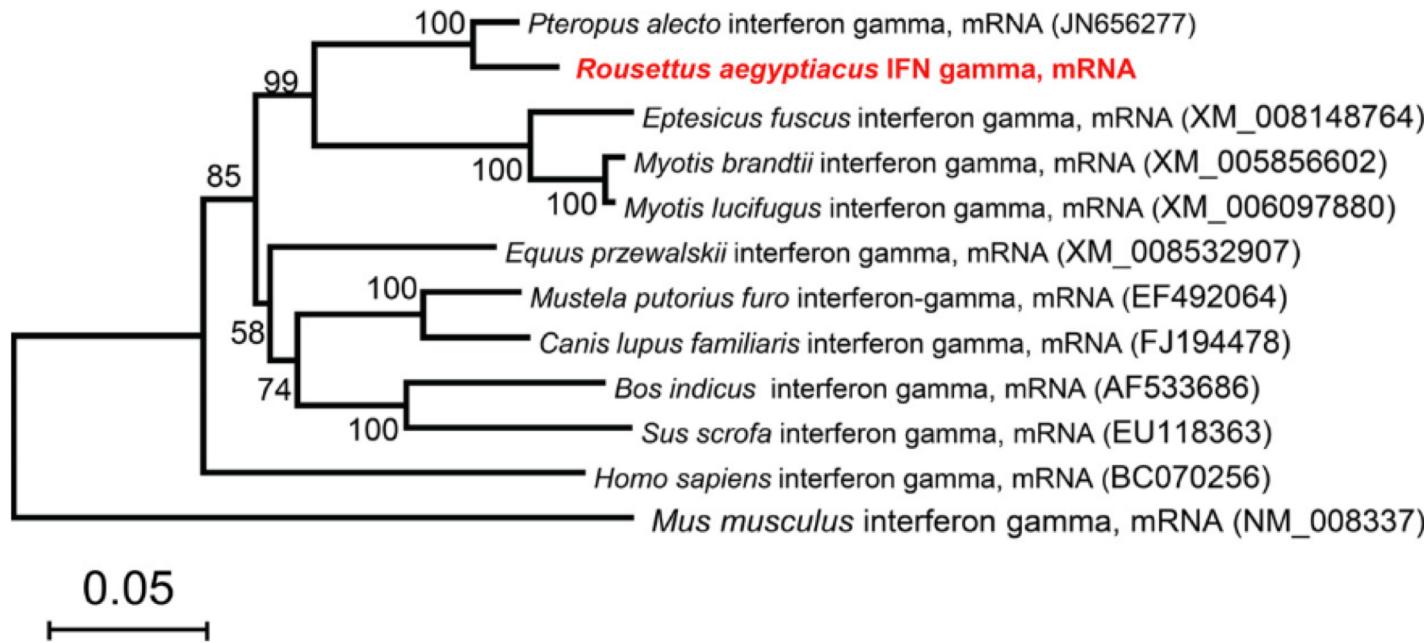
Table 1. Innate and adaptive immune systems in bats and birds

	Protein	Function	Bats	Birds
Innate immunity	RIG-I	Cytosolic PRR; detects dsRNA	Present and functional in the black flying fox (<i>Pteropus alecto</i> ; AEW46678); homolog also present in David's Myotis (<i>Myotis davidii</i> ; ELK34300) [16]	Present and functional in mallard duck (ACA61272) and present in goose (ADV58759), but absent in chicken [33]
	MDA5	Cytosolic PRR; detects dsRNA	Present and functional in black flying fox (<i>P. alecto</i> ; AEW46679); homolog also present in David's Myotis (<i>M. davidii</i> ; ELK28159)	Present in mallard duck (GU936632), goose (AGC51036), and chicken (ADD83027) [33,86]
	LGP2	Cytosolic PRR; detects dsRNA	Present and functional in black flying fox (<i>P. alecto</i>); homolog also present in David's Myotis (<i>M. davidii</i>)	Present in chicken [34]
	TLR7	Membrane-bound PRR; detects ssRNA	Present in black flying fox (<i>P. alecto</i> ; ADO01609), Leschenault's rousette (<i>Rousettus leschenaultia</i> ; BAH02556), and David's Myotis (<i>M. davidii</i> ; ELK30184) [15,85]	Present in chicken (ACR26250) and mallard duck (ABK51522) [35,87,88]
	TLR3	Membrane-bound PRR; detects dsRNA	Present in black flying fox (<i>P. alecto</i> ; ADO01605), Leschenault's rousette (<i>R. leschenaultia</i> ; BAH02555), and David's Myotis (<i>M. davidii</i> ; ELK23529) [15,85]	Present in chicken (ABG79022) and muscovy duck (AFK29094) [89]
	TLR8	Membrane-bound PRR; detects ssRNA	Present in black flying fox (<i>P. alecto</i> ; ELK17709) and David's Myotis (<i>M. davidii</i> ; ELK30183) [85]	Disrupted in chicken and duck genome [90]

Positive selection in immune genes

Toll like receptors in bats

- Toll-like receptors represent a largely evolutionarily conserved pathogen recognition machinery responsible
- First line of defense of the immune system



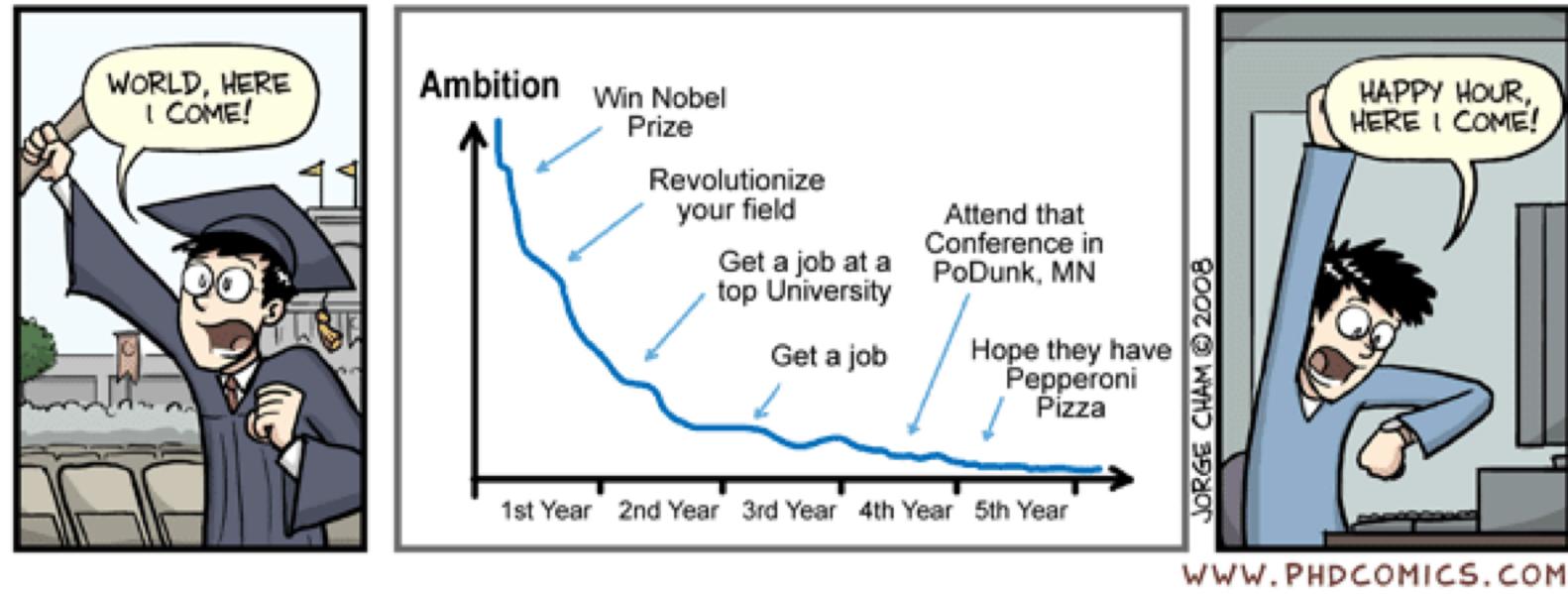
Chan et al. 2013

Aims

Investigate gene evolution of TLR proteins in a group of 36 bat species.

Study which set of genes are subject to positive selection in bats dN/dS).

YOUR LIFE AMBITION - What Happened??



↪ SUMAC 2.23: supermatrix constructor

SUMAC (Supermatrix Constructor) is a Python package to data-mine GenBank, construct phylogenetic supermatrices, and assess the phylogenetic decisiveness of a matrix given the pattern of missing sequence data. SUMAC calculates a novel metric, Missing Sequence Decisiveness Scores (MSDS), which measure how much each individual missing sequence contributes to the decisiveness of the matrix. MSDS can be used to compare supermatrices and prioritize the acquisition of new sequence data.

```
python -m sumac -d MAM -i Myotis -o Murina -minl 750 cores 4 -g  
/Users/diegoellis/projects/development/Comparative_genomics/finalproject/Data/TLR_gene_9_myotis_lucifugus.FASTA -de
```

```
Searching for ingroup and outgroup sequences...  
Ingroup sequences found: 8920 Outgroup sequences found: 1895 Percent searched: 100.0  
Using sequence length similarity threshold 0.25  
Using UCLUST id threshold 0.5  
Using BLAST e-value threshold 1e-10
```

1. Getting the sequences

NCBI Protein sequences

NCBI Resources How To

Protein TLR7 Myotis

Create alert Advanced

Sign in to NCBI Help

Species Animals (5) Customize ...

Source databases RefSeq (4) Customize ...

Sequence length Custom range...

Molecular weight Custom range...

Release date Custom range...

Revision date Custom range...

Items: 5

toll-like receptor 7 [Myotis lucifugus]
1. 1044 aa protein
Accession: XP_023609502.1 GI: 1335132425
BioProject Nucleotide Taxonomy
GenPept Identical Proteins FASTA Graphics

PREDICTED: LOW QUALITY PROTEIN: toll-like receptor 7 [Myotis davidii]
2. 1074 aa protein
Accession: XP_015419449.1 GI: 987945870
BioProject Nucleotide Taxonomy
GenPept Identical Proteins FASTA Graphics

PREDICTED: toll-like receptor 7 [Myotis brandtii]
3. 1059 aa protein
Accession: XP_005881008.2 GI: 946797482
BioProject Nucleotide Taxonomy
GenPept Identical Proteins FASTA Graphics

PREDICTED: toll-like receptor 7 [Myotis brandtii]
4. 1059 aa protein
Accession: XP_014384420.1 GI: 946797480
BioProject Nucleotide Taxonomy
GenPept Identical Proteins FASTA Graphics

toll-like receptor 7 [Myotis ricketti]

Summary Sort by Default order Send to: Filters: Manage Filters

See TLR7 toll like receptor 7 in the Gene database
tlr7 reference sequences Transcript (2) Protein (2)

Results by taxon

Top Organisms [Tree]
Myotis brandtii (2)
Myotis davidii (1)
Myotis ricketti (1)
Myotis lucifugus (1)

Analyze these sequences

Run BLAST

Align sequences with COBALT

Identify Conserved Domains with CD-Search

Find in these sequences

Find related data

Database: Select

Find items

Search details

TLR7[All Fields] AND ("Myotis" [Organism] OR Myotis[All Fields])

Search See more...



1. Getting the sequences

NIH U.S. National Library of Medicine NCBI National Center for Biotechnology Information Sign in to NCBI

BLAST® » blastp suite Home Recent Results Saved Strategies Help

Standard Protein BLAST

Enter Query Sequence

Enter accession number(s), gi(s), or FASTA sequence(s) SISAALFLMVMTASHLYFWDVVWYSYHFCKAKIKGYQRLVSPDSCYDAFIYDTKDPAVTEWLDE
LVAR
LEDPREKHFNLCLEERDWLPGQPVLNLTQSILSKKTVFVMTDKYAKTENFKIAFYLSHQRLIDEK
VDV
IILIFLEKPLQSKFLQLRKRLCGSSVLEWPTNPQAHPFWQCLKNALATDNHVTYSRKFETV

Query subrange
From
To

We are beta testing a New Results page Click here if you would like to see your results in the new format. You can always switch back to the Traditional Results page.

Choose Search Set

Database Non-redundant protein sequences (nr)

Organism Optional Enter organism name or id--completions will be suggested exclude Enter organism common name, binomial, or tax id. Only 20 top taxa will be shown.

Exclude Optional Models (XM/XP) Non-redundant RefSeq proteins (WP) Uncultured/environmental sample sequences

Entrez Query Optional YouTube Create custom database Enter an Entrez query to limit search

Program Selection

Algorithm Quick BLASTP (Accelerated protein-protein BLAST)
 blastp (protein-protein BLAST)
 PSI-BLAST (Position-Specific Iterated BLAST)
 PHI-BLAST (Pattern Hit Initiated BLAST)
 DELTA-BLAST (Domain Enhanced Lookup Time Accelerated BLAST)

Choose a BLAST algorithm

1. Getting the sequences

Sequences producing significant alignments:

Select: All None Selected:0

	Description	Max Score	Total Score	Query Cover	E value	Per. Ident	Accession
<input type="checkbox"/>	toll-like receptor 7 [Myotis lucifugus]	2136	2136	100%	0.0	100.00%	XP_023609502.1
<input type="checkbox"/>	PREDICTED: toll-like receptor 7 [Myotis brandtii]	2127	2127	100%	0.0	99.52%	XP_014384420.1
<input type="checkbox"/>	Toll-like receptor 7 [Myotis brandtii]	2124	2124	100%	0.0	99.52%	EPQ16883.1
<input type="checkbox"/>	toll-like receptor 7 [Myotis ricketti]	2108	2108	100%	0.0	98.66%	APR72762.1
<input type="checkbox"/>	PREDICTED: LOW QUALITY PROTEIN: toll-like receptor 7 [Myotis davidii]	2100	2100	100%	0.0	98.18%	XP_015419449.1
<input type="checkbox"/>	Toll-like receptor 7 [Myotis davidii]	2097	2097	100%	0.0	98.18%	ELK30184.1
<input type="checkbox"/>	toll-like receptor 7 [la io]	1997	1997	100%	0.0	95.98%	APR72766.1
<input type="checkbox"/>	toll-like receptor 7 [Eptesicus fuscus]	1994	1994	100%	0.0	95.69%	XP_008154799.1
<input type="checkbox"/>	PREDICTED: toll-like receptor 7 [Miniopterus natalensis]	1860	1860	99%	0.0	88.30%	XP_016071121.1
<input type="checkbox"/>	toll-like receptor 7 [Miniopterus schreibersii]	1859	1859	99%	0.0	88.21%	APR72768.1
<input type="checkbox"/>	PREDICTED: toll-like receptor 7 isoform X1 [Rousettus aegyptiacus]	1833	1833	100%	0.0	86.79%	XP_015975065.1
<input type="checkbox"/>	PREDICTED: toll-like receptor 7 isoform X2 [Rousettus aegyptiacus]	1833	1833	100%	0.0	86.79%	XP_015975073.1
<input type="checkbox"/>	toll-like receptor 7 [Rhinolophus sinicus]	1832	1832	100%	0.0	86.03%	APR72764.1
<input type="checkbox"/>	toll-like receptor 7 [Pteropus vampyrus]	1831	1831	100%	0.0	86.51%	XP_011361212.1
<input type="checkbox"/>	toll-like receptor 7 [Eonycteris spelaea]	1824	1824	100%	0.0	86.22%	APR72765.1
<input type="checkbox"/>	PREDICTED: toll-like receptor 7 isoform X2 [Ceratotherium simum simum]	1823	1823	100%	0.0	86.62%	XP_004435171.1
<input type="checkbox"/>	PREDICTED: toll-like receptor 7 isoform X1 [Ceratotherium simum simum]	1821	1821	100%	0.0	86.62%	XP_014646590.1
<input type="checkbox"/>	PREDICTED: toll-like receptor 7 [Galeopterus variegatus]	1818	1818	100%	0.0	85.37%	XP_008569554.1
<input type="checkbox"/>	toll-like receptor 7 [Rhinolophus affinis]	1814	1814	100%	0.0	85.93%	APR72763.1
<input type="checkbox"/>	toll-like receptor 7 isoform X1 [Desmodus rotundus]	1810	1810	100%	0.0	86.70%	XP_024425637.1

1. Getting the sequences - Outgroup

Protein

Protein Help

Create alert Advanced

Species
Animals (744)
Plants (1)
Bacteria (3)
Viruses (2)
Customize ...

Source databases
PDB (244)
RefSeq (152)
UniProtKB / Swiss-Prot (37)
Customize ...

Sequence length
Custom range...

Molecular weight
Custom range...

Release date
Custom range...

Revision date
Custom range...

[Clear all](#)

[Show additional filters](#)

Summary ▾ 20 per page ▾ Sort by Default order ▾

Send to: ▾ Filters: [Manage Filters](#)

See the [results of this search \(20 items\)](#) in our new [Identical Protein Groups](#) database.

Items: 1 to 20 of 750

Selected: 2

<< First < Prev Page 1 of 38 Next > Last >>

[interleukin 1 receptor precursor \[Homo sapiens\]](#)
1. 569 aa protein
Accession: AAA59137.1 GI: 307046
[Nucleotide](#) [PubMed](#) [Taxonomy](#)
[GenPept](#) [Identical Proteins](#) [FASTA](#) [Graphics](#)

[interleukin 1 receptor, type II, isoform CRA_a \[Homo sapiens\]](#)
2. 398 aa protein
Accession: EAX01803.1 GI: 119622208
[BioProject](#) [Nucleotide](#) [PubMed](#) [Taxonomy](#)
[GenPept](#) [Identical Proteins](#) [FASTA](#) [Graphics](#)

[interleukin 1 receptor, type II, isoform CRA_a \[Homo sapiens\]](#)
3. 398 aa protein
Accession: EAX01802.1 GI: 119622207
[BioProject](#) [Nucleotide](#) [PubMed](#) [Taxonomy](#)
[GenPept](#) [Identical Proteins](#) [FASTA](#) [Graphics](#)

[interleukin 1 receptor, type II, isoform CRA_a \[Homo sapiens\]](#)

Results by taxon

Top Organisms [\[Tree\]](#)
[Homo sapiens \(699\)](#)
[Cervus elaphus hippelaphus \(12\)](#)
[Ophiophagus hannah \(9\)](#)
[Colinus virginianus \(5\)](#)
[Callipepla squamata \(4\)](#)
[All other taxa \(21\)](#)
[More...](#)

Find related data

Database: [Select](#)

[Find items](#)

Search details

("Homo sapiens" [Organism] OR Homo sapiens [All Fields]) AND interleukin 1 receptor [All Fields]

See more...

Choose Destination

File Clipboard Collections

[Homo sapiens]

Download 1 item.

Format [FASTA](#)

Sort by Default order

Show GI

[Create File](#)

2. Alignment

MAFFT version 7

Multiple alignment program for amino acid or nucleotide sequences

```
MEDYTKIEKIGEG-----TYGVVYKGRHKTTGQVVAMKKIRLESEEEGVPSTAIRE
MEDYTKIEKIGEG-----TYGVVYKGRHKTTGQVVAMKKIRLESEEEGVPSTAIRE
MEDYTKIEKIGEG-----TYGVVYKGRHKTTGQVVAMKKIRLESEEEGVPSTAIRE
MEDYTKIEKIGEGEWLYYYTHN-----HKTTGQVVAMKKIRLESEEEGVPSTAIRE
MEDYTKIEKIGEG-----TYGVVYKGRHKTTGQIVAMKKIRLESEEEGVPSTAIRE
MDDYIKIEKIGEG-----TYGVVYKGRHKTTGQIVAMKKIRLESEEEGVPSTAIRE
-----TYGVVYKGRHRATGQIVAMKKIRLESEEEGVPSTAIRE
MEDYIKIEKIGEG-----TYGVVYKGRHKTTGQIVAMKKIRLESEEEGVPSTAIRE
```

3. Trimming

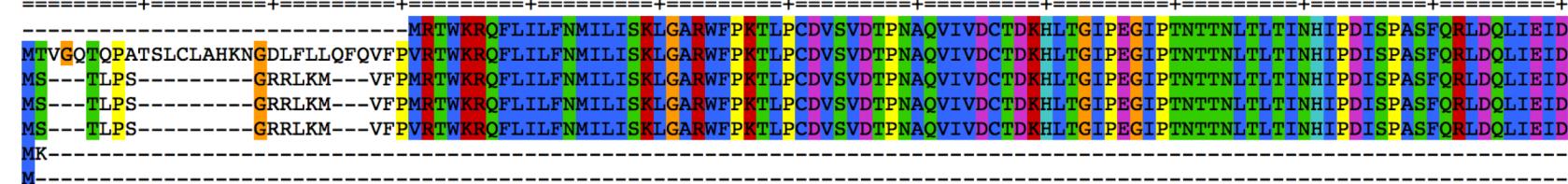


Selected Sequences: 7 /Selected Residues: 305
Deleted Sequences: 0 /Deleted Residues: 868

Gaps Scores:

=0= <.001 <.050 <.100 <.150 <.200 <.250 <.350 <.500 <.750 <1.00 =1=

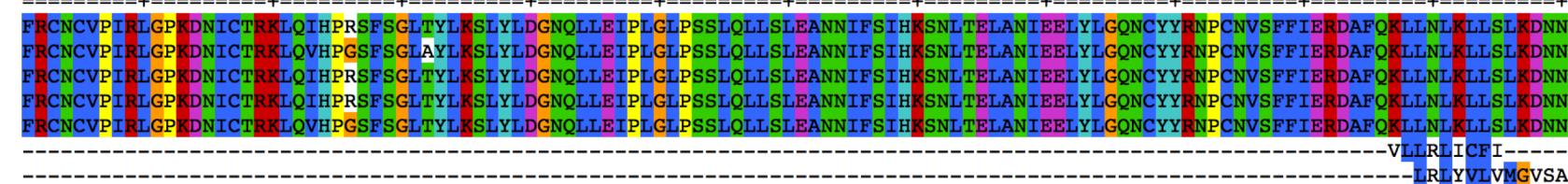
10 20 30 40 50 60 70 80 90 100 110 120



Selected Cols:

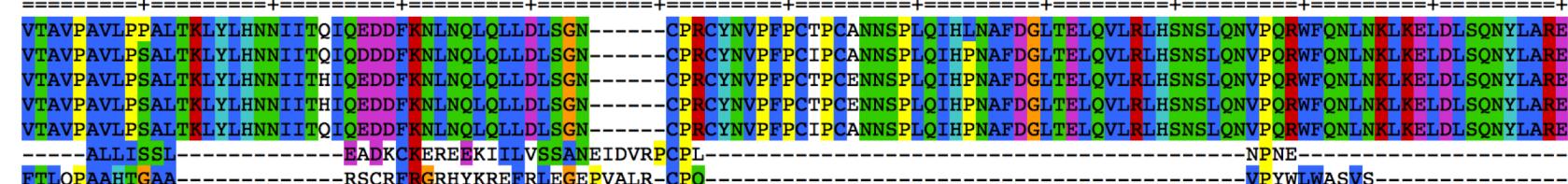
Gaps Scores:

130 140 150 160 170 180 190 200 210 220 230 240



Gaps Scores:

250 260 270 280 290 300 310 320 330 340 350 360



Gaps Scores:

4 Building phylogenetic gene tree

IQ-TREE

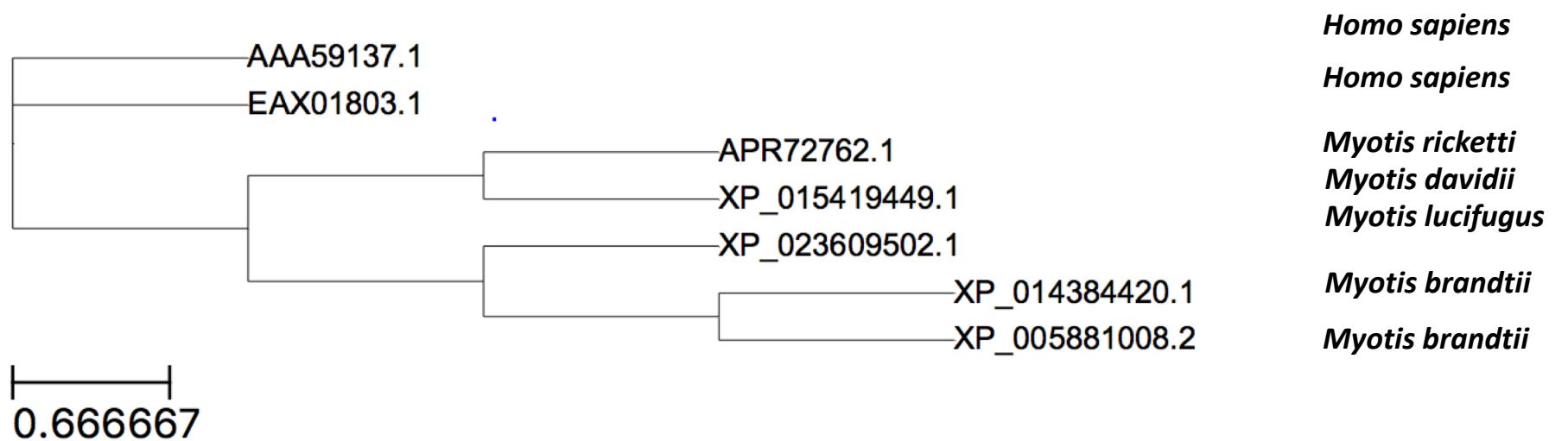
Efficient software for phylogenomic inference

```
Creating bootstrap support values...
Tree with assigned bootstrap support written to trimed_inout_TLR.fasta.treefile
Analysis results written to:
  IQ-TREE report:          trimed_inout_TLR.fasta.iqtree
  Maximum-likelihood tree: trimed_inout_TLR.fasta.treefile
  Likelihood distances:    trimed_inout_TLR.fasta.mldist
  Screen log file:         trimed_inout_TLR.fasta.log
Total CPU time for bootstrap: 9241.766 seconds.
Total wall-clock time for bootstrap: 9305.960 seconds.

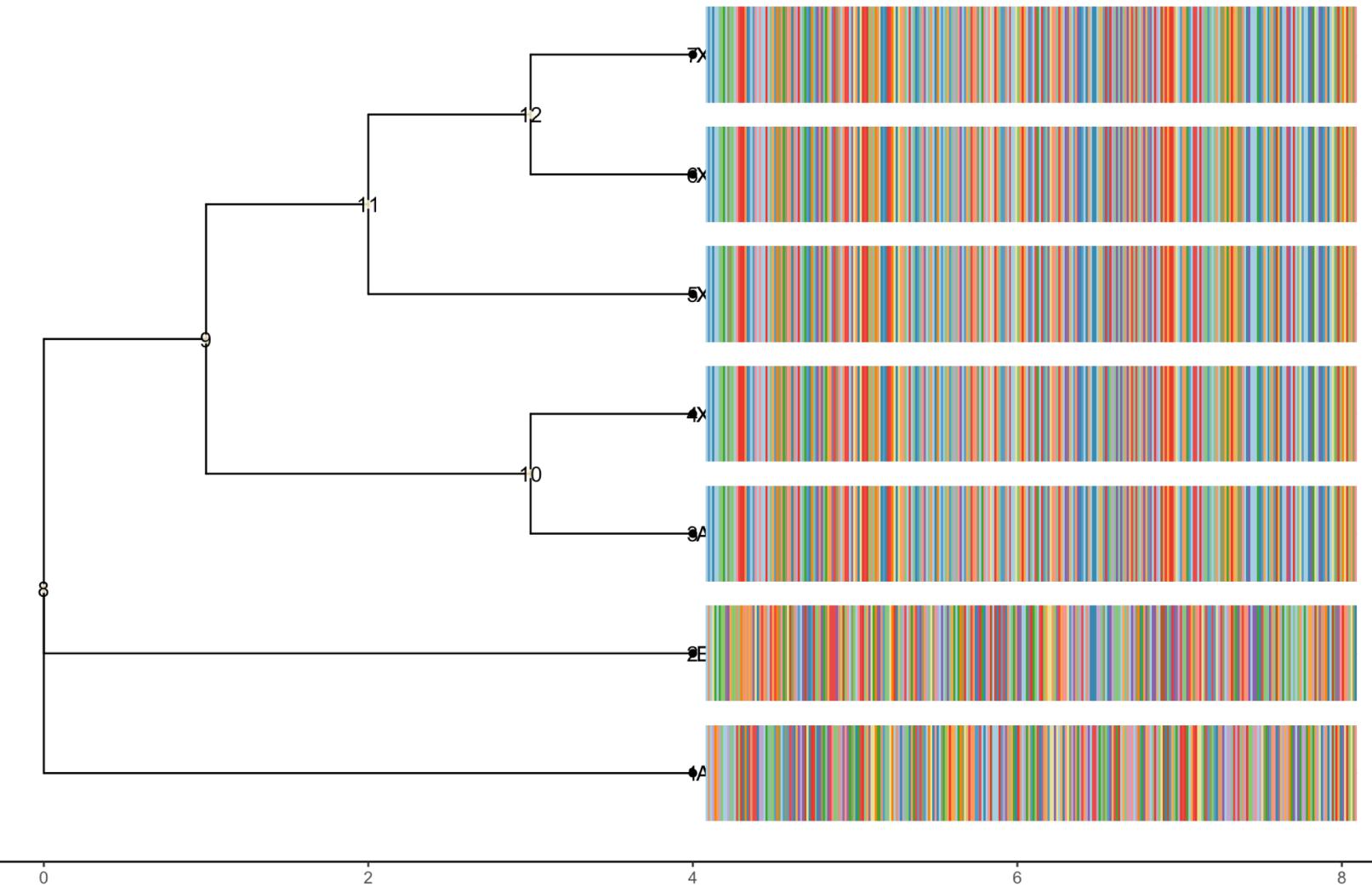
Non-parametric bootstrap results written to:
  Bootstrap trees:          trimed_inout_TLR.fasta.boottrees
  Consensus tree:           trimed_inout_TLR.fasta.contree
```

- R::ggtree and phyon ete3 for visualization

5 Tree visualization –TLR 7

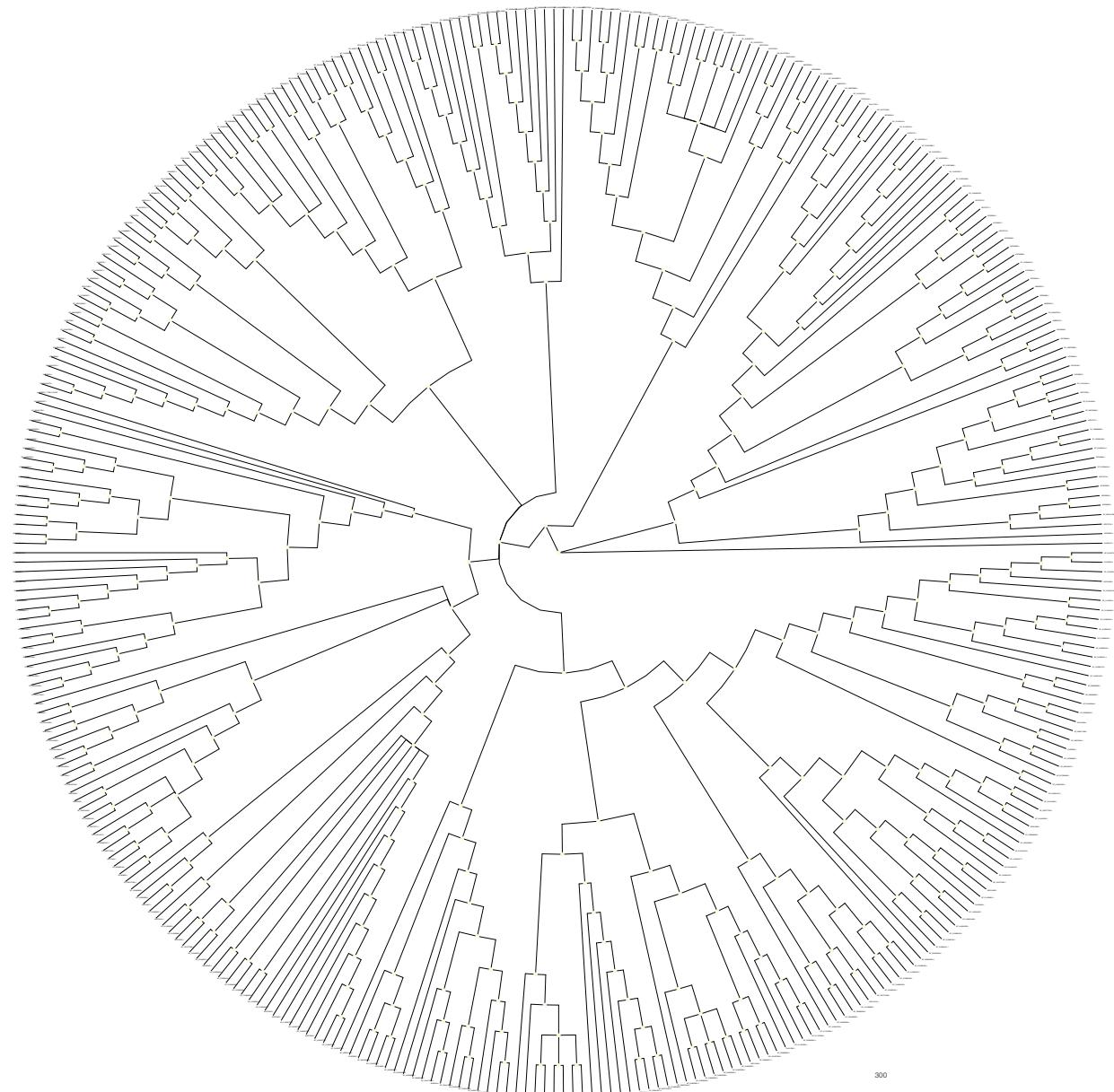


5 Tree visualization –TLR 7

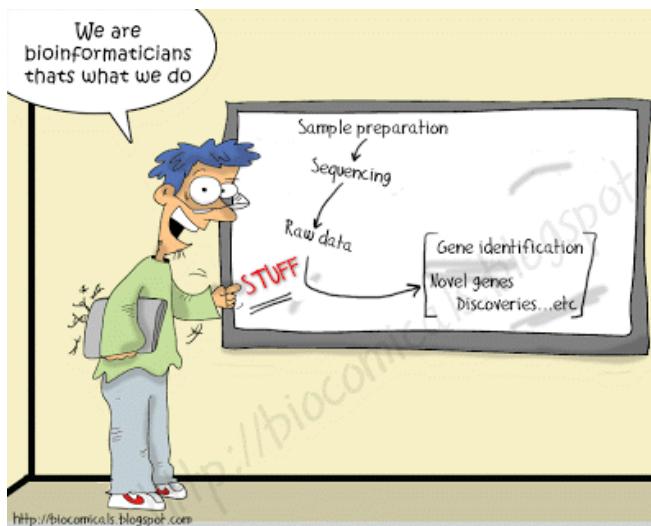


Phylogenetics of TLR genes in the *Chiroptera* genus

- 36 species, XXX families
- XXX sequences



Conclusions



Bioinformatics is the Wild west

Spent most of the time understanding pipelines and getting things to work

M. davidii, M. ricketti, M. lucifugus more similar TLR 7 than M. brandtii

Should have looked at TLR7 in all *Chiroptera* species

Next steps

CAFE: a computational tool for the study of gene family evolution 

Tijl De Bie, Nello Cristianini, Jeffery P. Demuth, Matthew W. Hahn ✉

Bioinformatics, Volume 22, Issue 10, 15 May 2006, Pages 1269–1271,

<https://doi.org/10.1093/bioinformatics/btl097>

Published: 16 March 2006 Article history ▾

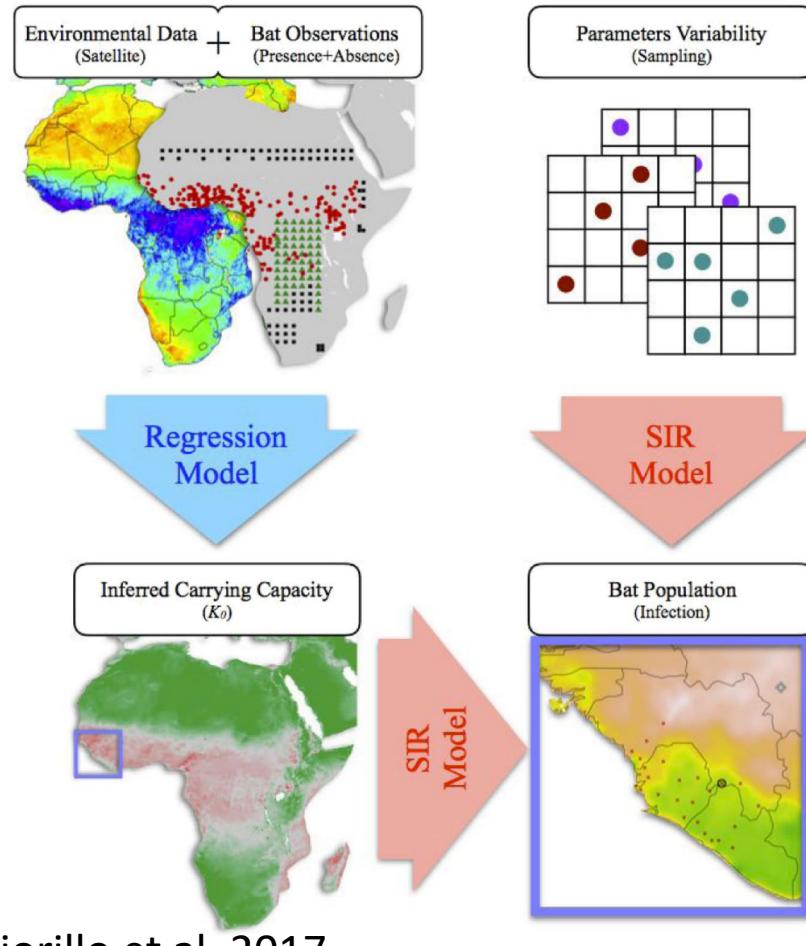
Contingency and Determinism in Replicated Adaptive Radiations of Island Lizards

Jonathan B. Losos,* Todd R. Jackman, Allan Larson, Kevin de Queiroz, Lourdes Rodríguez-Schettino

1998

Next steps

Front. Ecol. Evol., 04 December 2018 | <https://doi.org/10.3389/fevo.2018.00208>



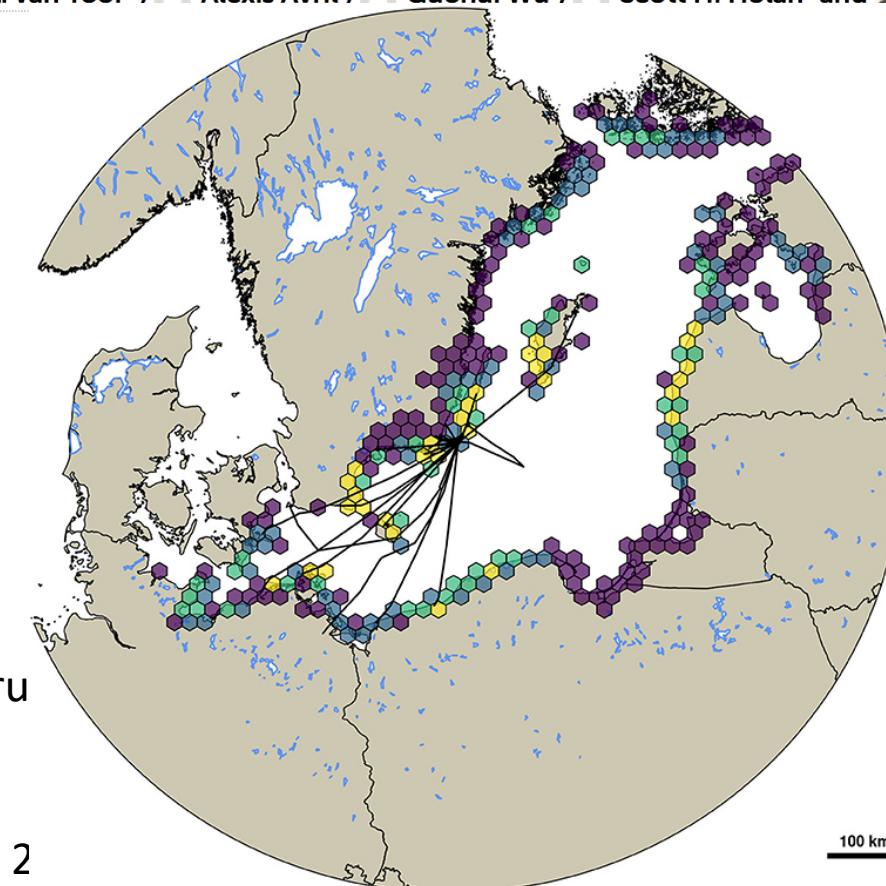
Fiorillo et al. 2017

Model Virome heterogeneity of ebola and connectivity in flying fru
(*Eidolon vesuvium*)

Similar work down on waterfowl (Wille et al. 2019, van Toor et al. 2

As the Duck Flies—Estimating the Dispersal of Low-Pathogenic Avian Influenza Viruses by Migrating Mallards

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Low risk area
Medium risk area
High risk area
Highest risk area

Well resolved genetic screening and evolutionary dynamics of Ebola measured

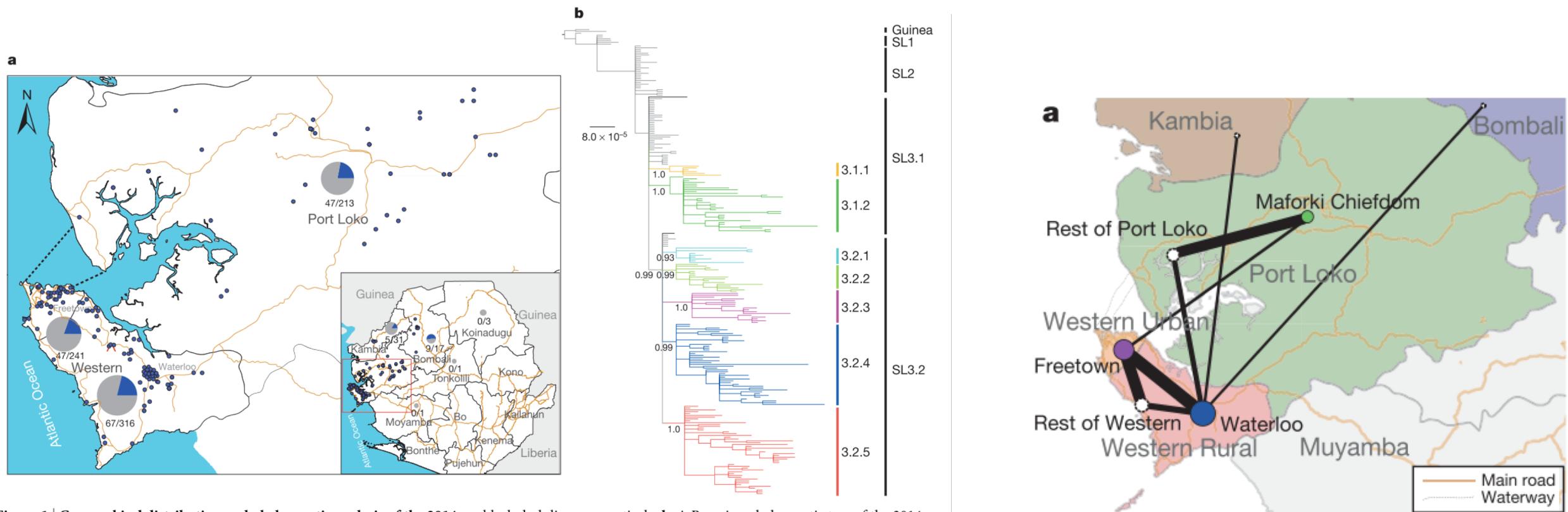


Figure 1 | Geographical distribution and phylogenetic analysis of the 2014 EBOV from Sierra Leone. a, Geographical distribution of the 823 EBOV positive samples and the 175 newly sequenced genomes (represented as blue dots). In the panel, main roads and waterways are showed as yellow lines and

black dash lines, respectively. b, A Bayesian phylogenetic tree of the 2014 EBOV. The 175 newly sequenced viruses in this study are shown in colours, and others are shown in grey. The seven novel lineages designated in the present are highlighted. Posterior support for major nodes is shown.

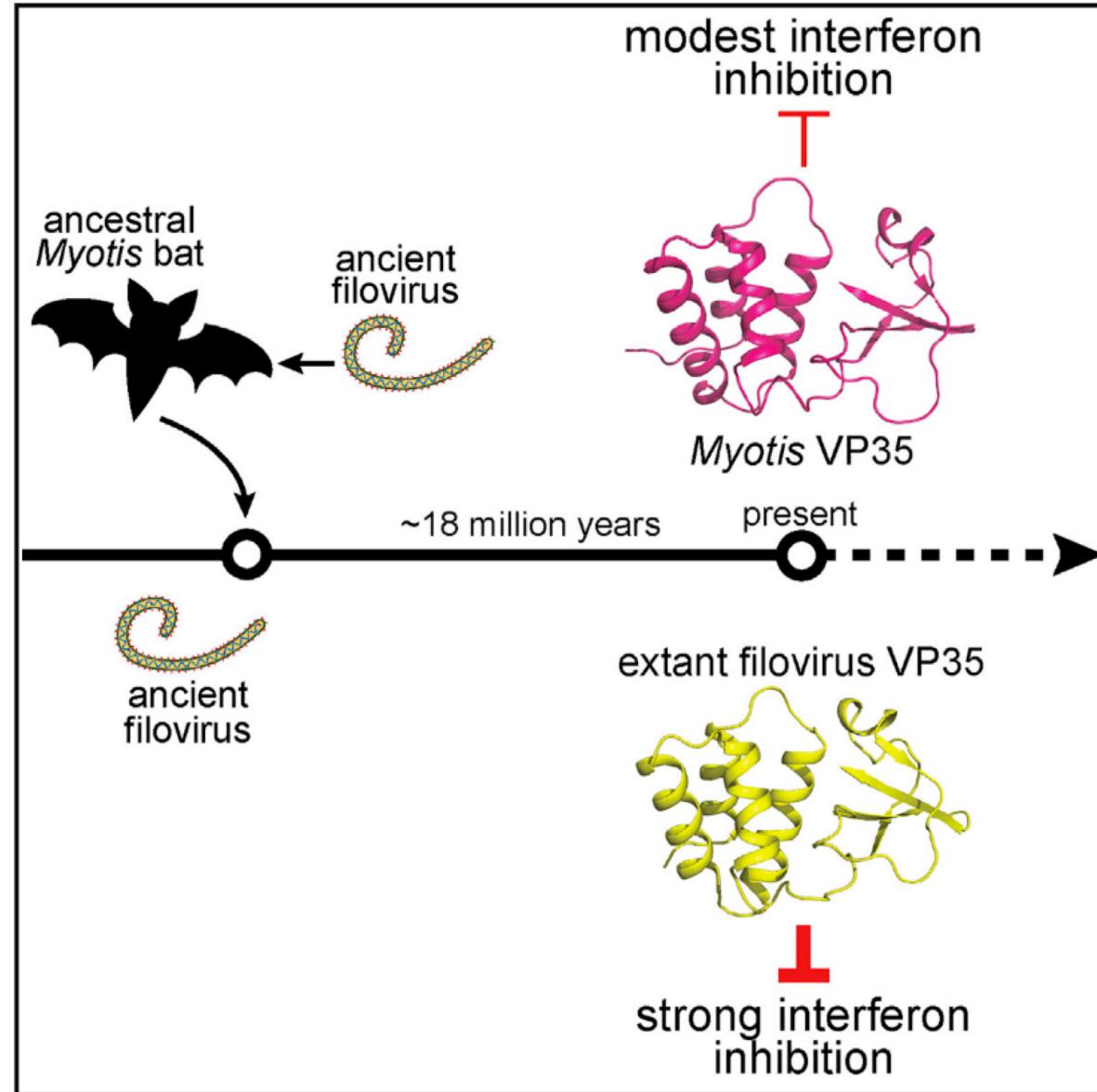
Thanks!



Antiviral Immunity of bats

- Immunity of bats heavy research interest
- Novel modes of antiviral defense (Pavlovich et al. 2018)

Graphical Abstract





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Next step

- Model Virome heterogeneity of ebola virus in bats (*Eidolon vesuvium*)

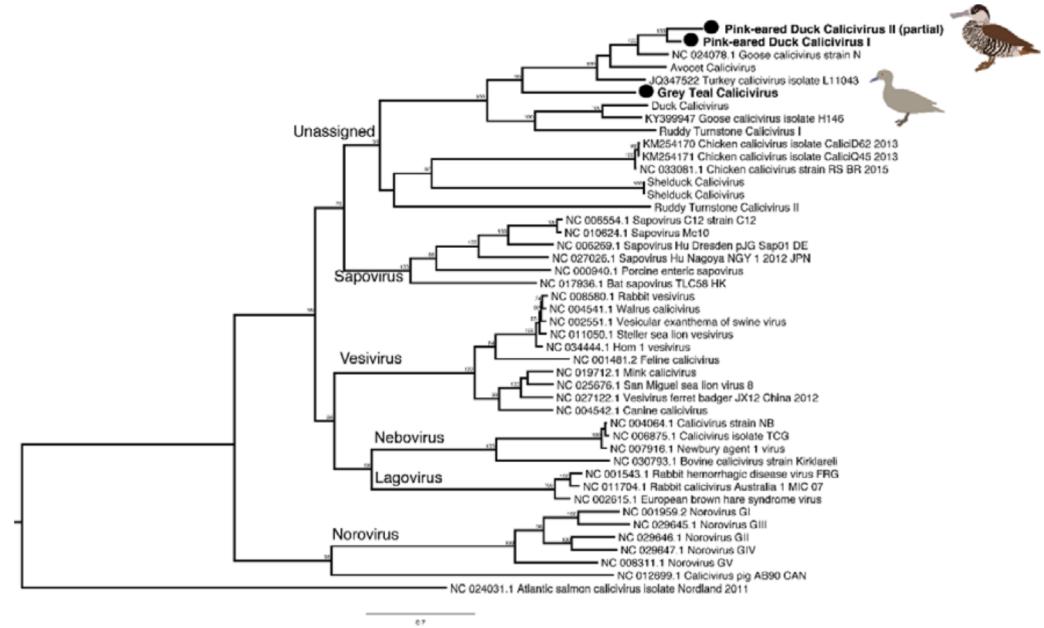


Figure 3. Phylogenetic tree of the virus polyprotein, including the RdRp, of representatives of the vertebrate RNA virus family the *Caliciviridae*. Viruses identified in this study are denoted with a filled circle and in bold. The most divergent calicivirus, Atlantic Salmon calicivirus, was used as

- Similar work has been performed in waterfowl (Wille et al. 2019, van Toor et al. in prep)