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# Real-time phylodynamics



Nuno Rodrigues Faria, on behalf of the ZiBRA team

Sir Henry Dale Fellow, Royal Society and Wellcome Trust

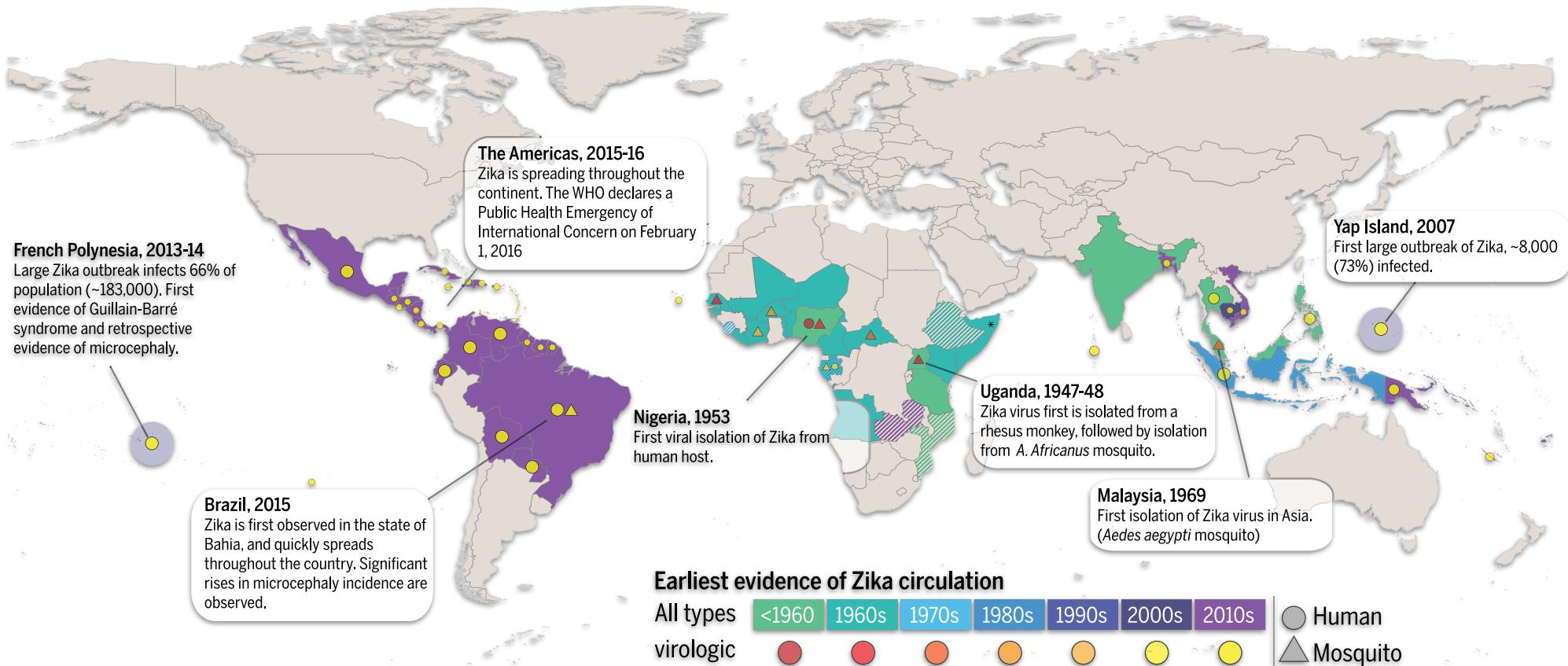
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Part 1

# Epidemiology of Zika virus

# History of Zika virus emergence from a serologic perspective

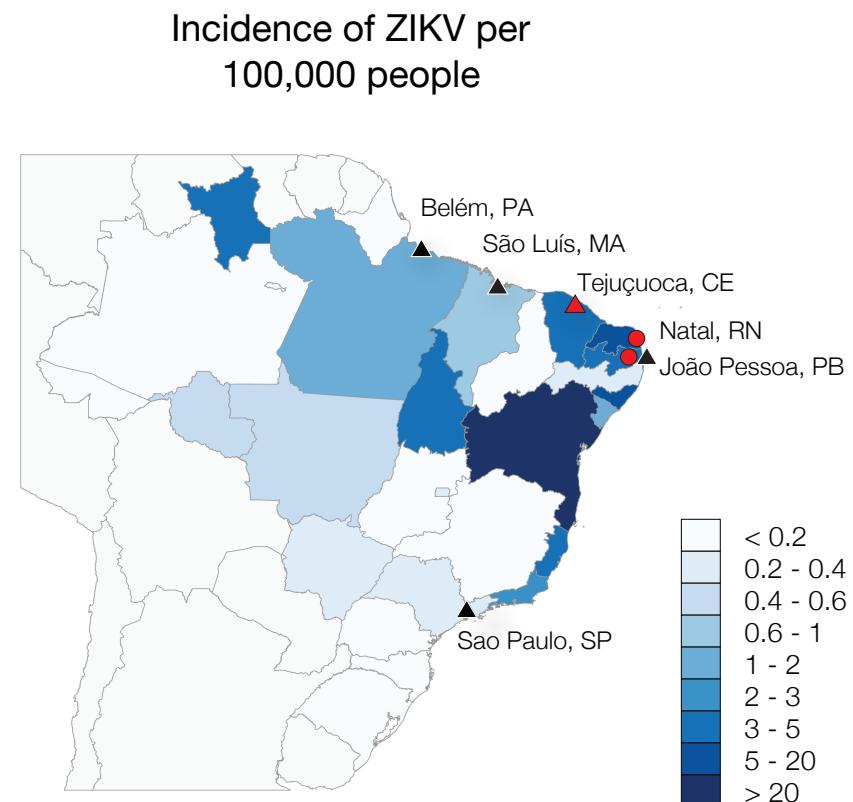
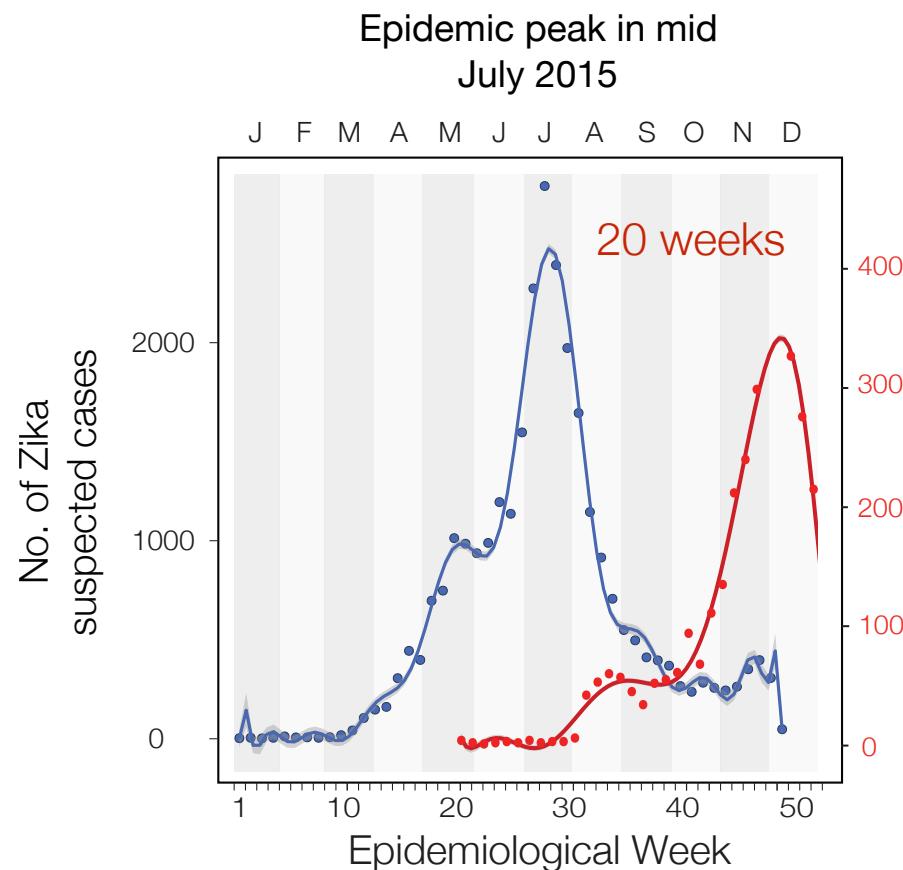


Lesller et al. Science Aug 2016

# Temporal association between ZIKV notified cases in 2015 and microcephaly

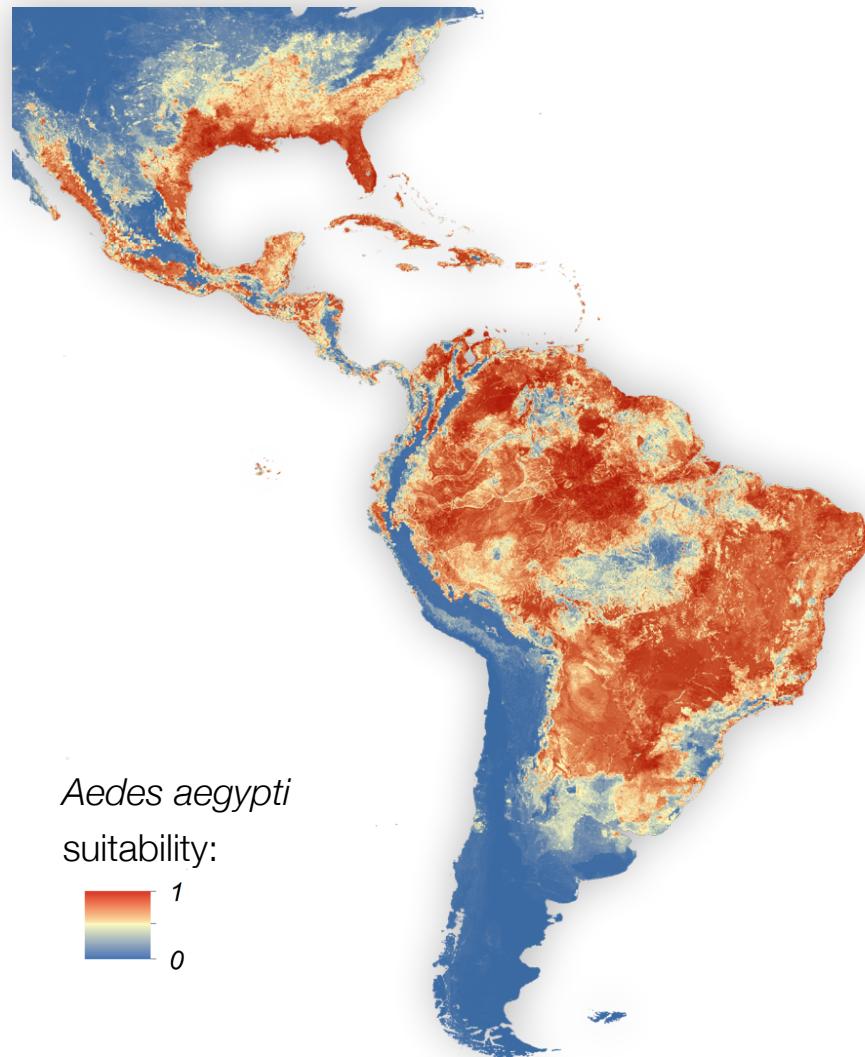
## Local transmission in Brazil: 07 May 2015

WHO Public Health Emergency of International Concern: 01 Feb 2016  
WHO/CDC confirm association between ZIKV infection and **microcephaly**: 13 Apr 2016

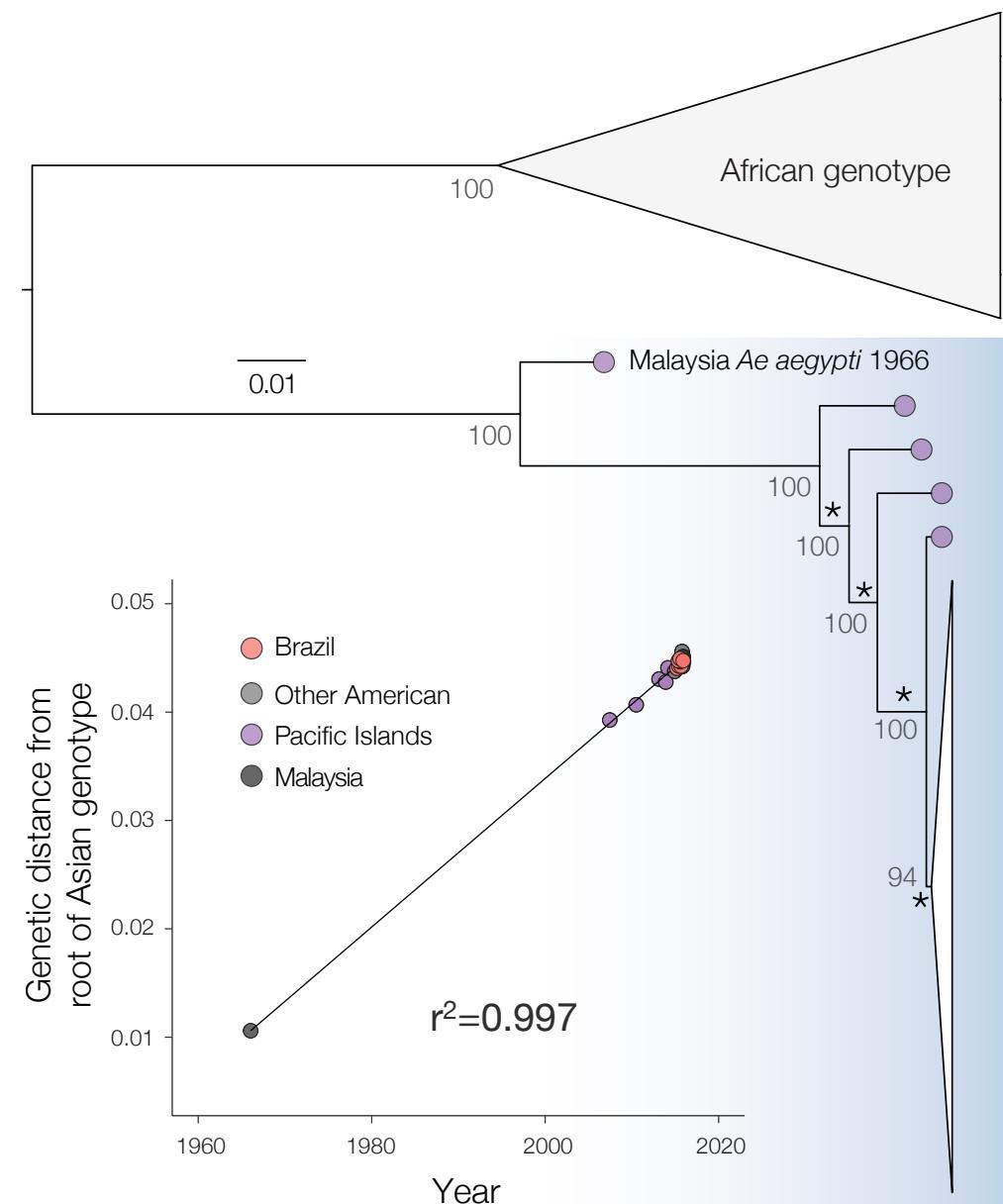


Faria et al. *Science* March 2016

# Emergence of ZIKV as a function of local transmission regime, viral pathogenesis



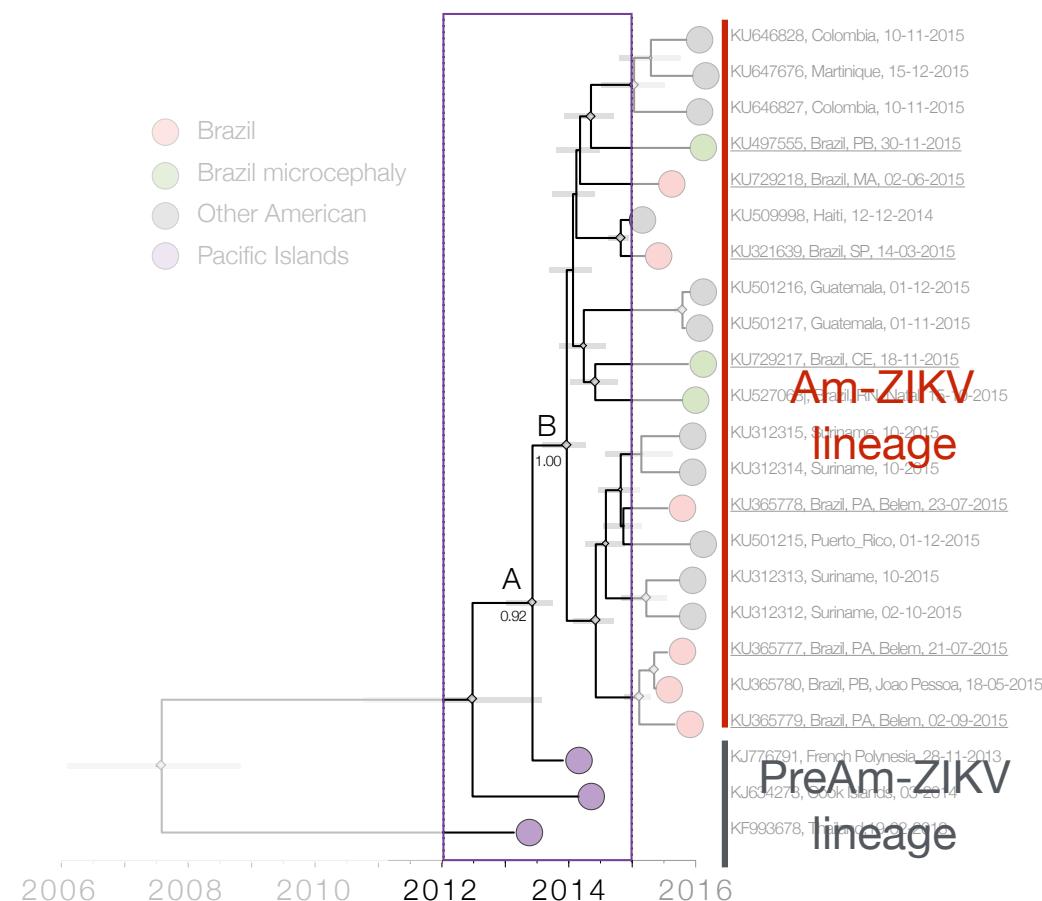
Kraemer MUG et al. eLife 2014



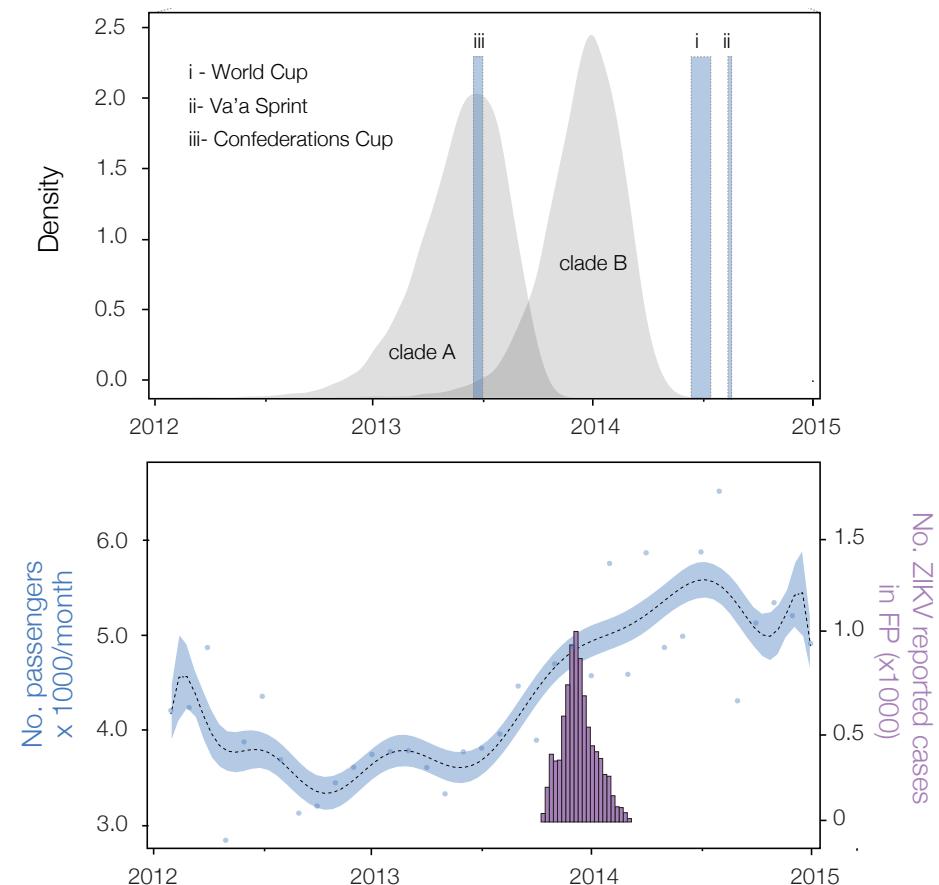
Faria et al. Science March 2016

# Emergence of ZIKV as a function of local transmission regime, viral pathogenesis and **global mobility**

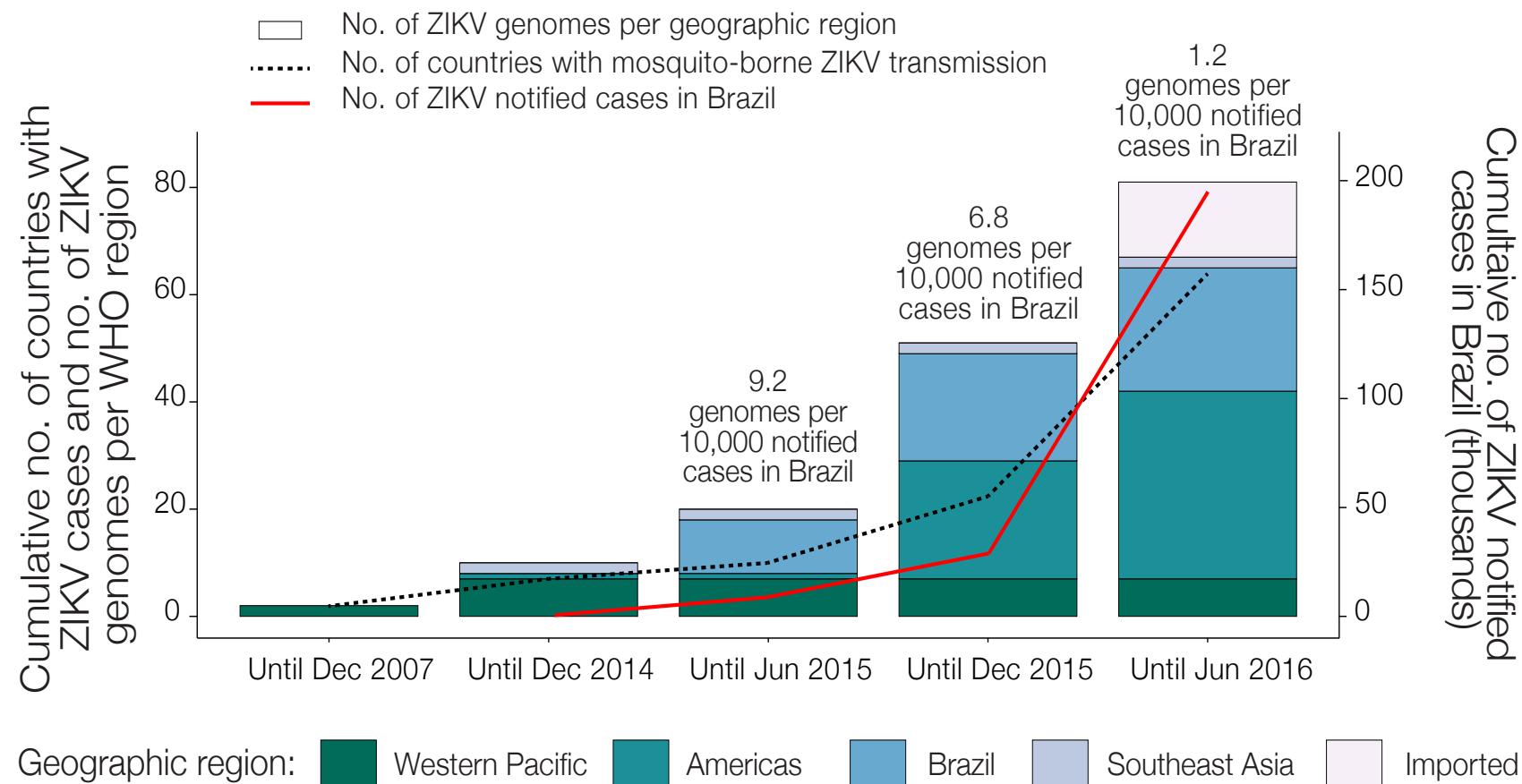
ZIKV Asian genotype  
molecular clock phylogeny



Combination of genetic  
and flight data

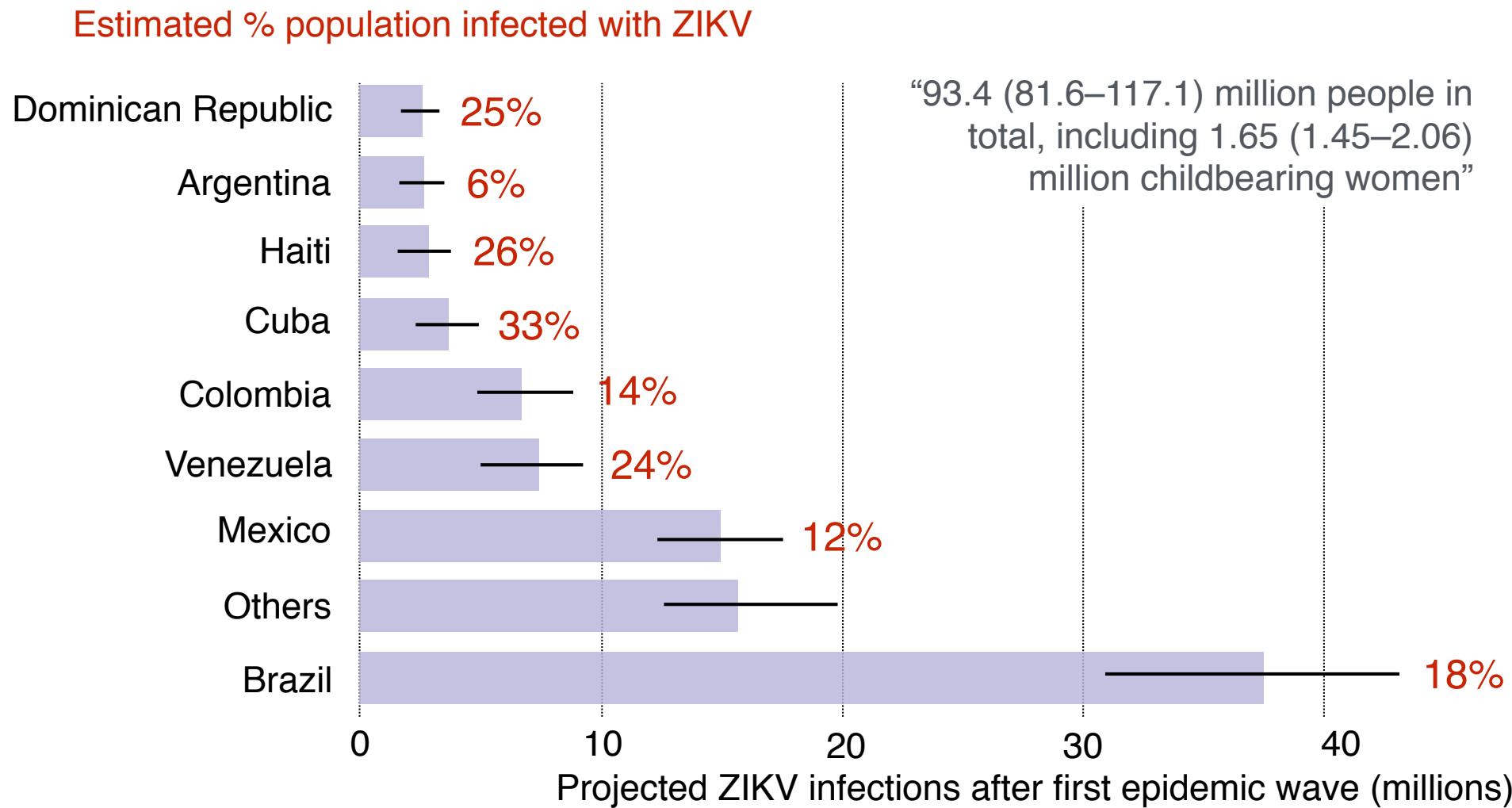


# 76 countries reporting ZIKV transmission but only 61 published genomes (as of Feb 2017)



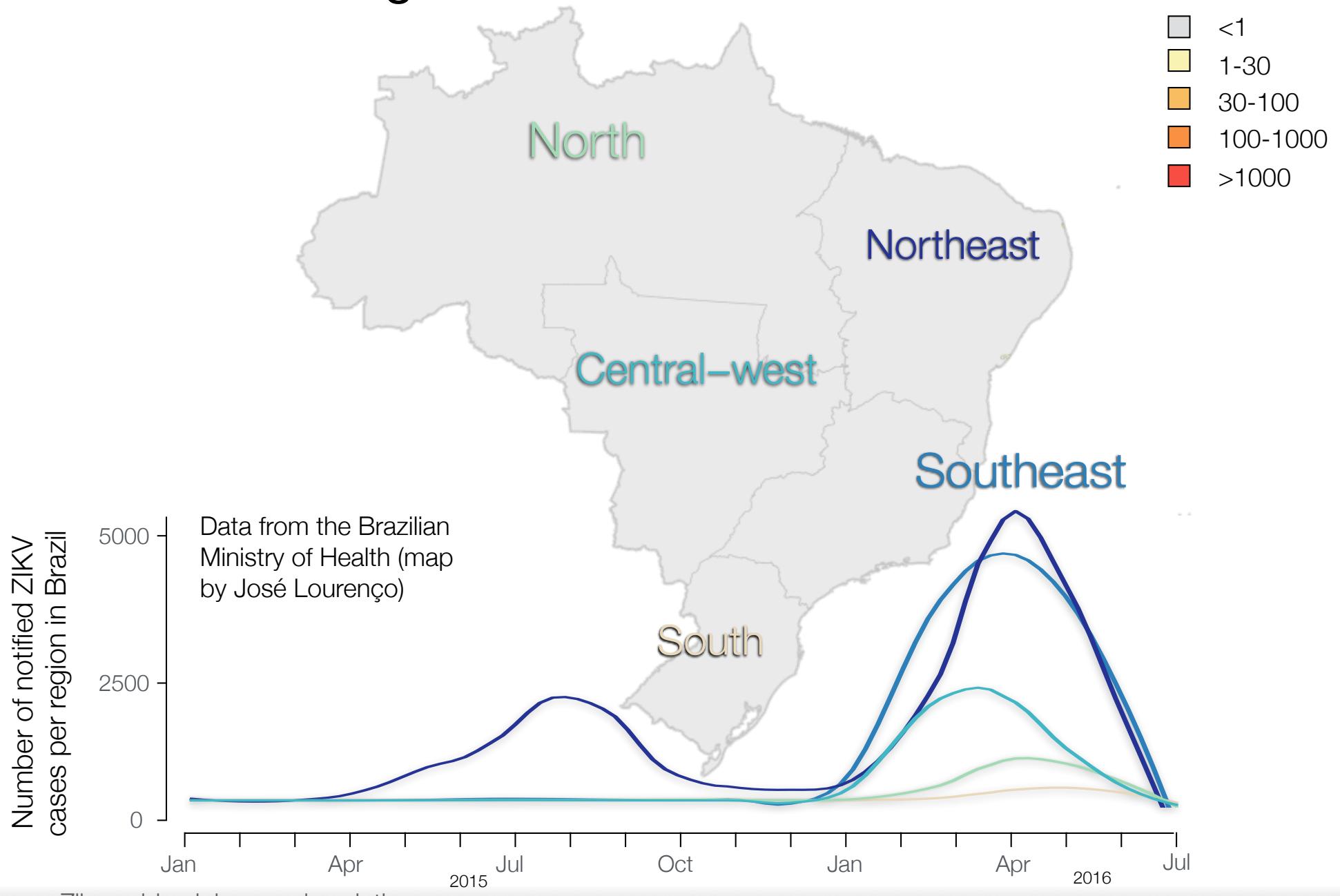
Faria et al, *Genome Medicine*, Sep 2016

# Estimates of the number of ZIKV infections in Brazil suggest around 37 million cases

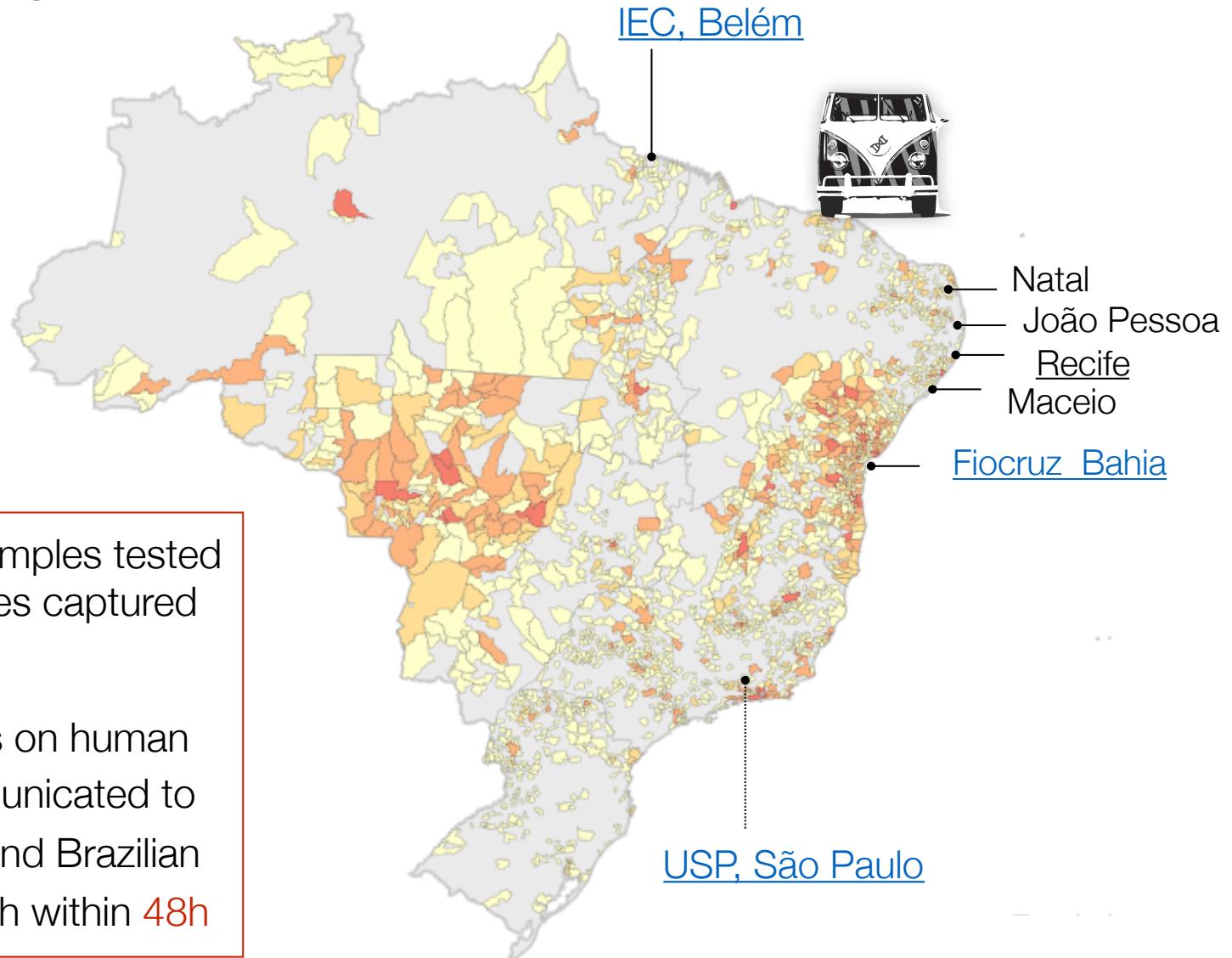


Perkins et al. *Nature Microb* Jul 2016

# Earliest ZIKV cases were in the Northeast region



# The ZiBRA project, a sequencing journey through ZIKV hotspots



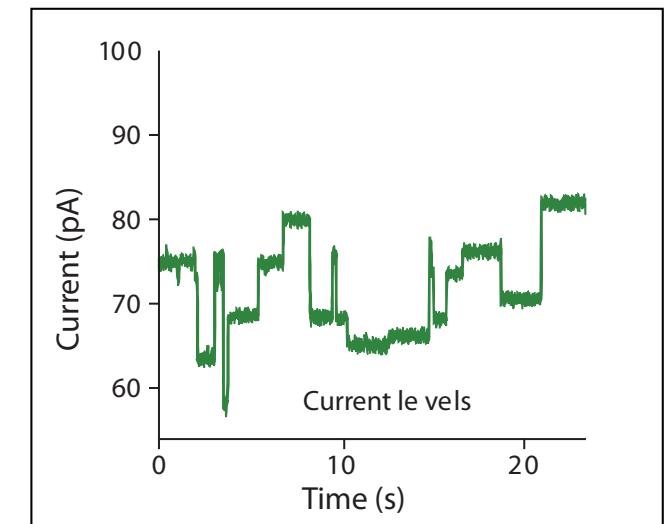
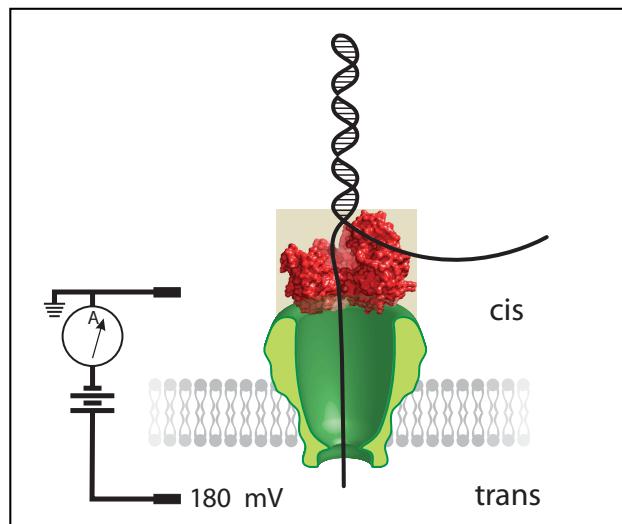
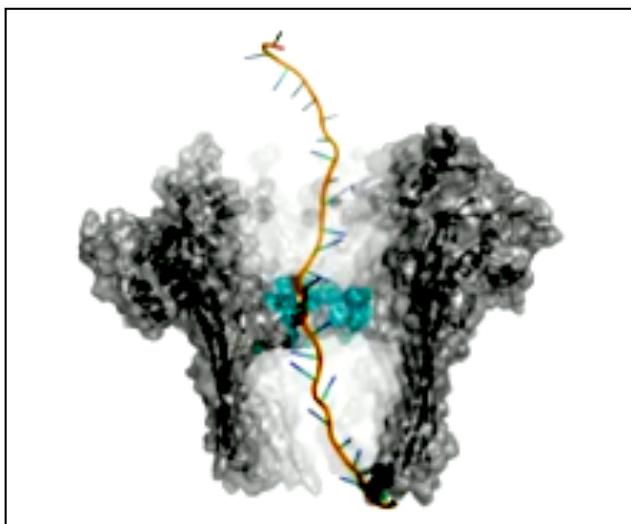
After the ZiBRA journey, molecular surveillance continued in SP and Bahia.



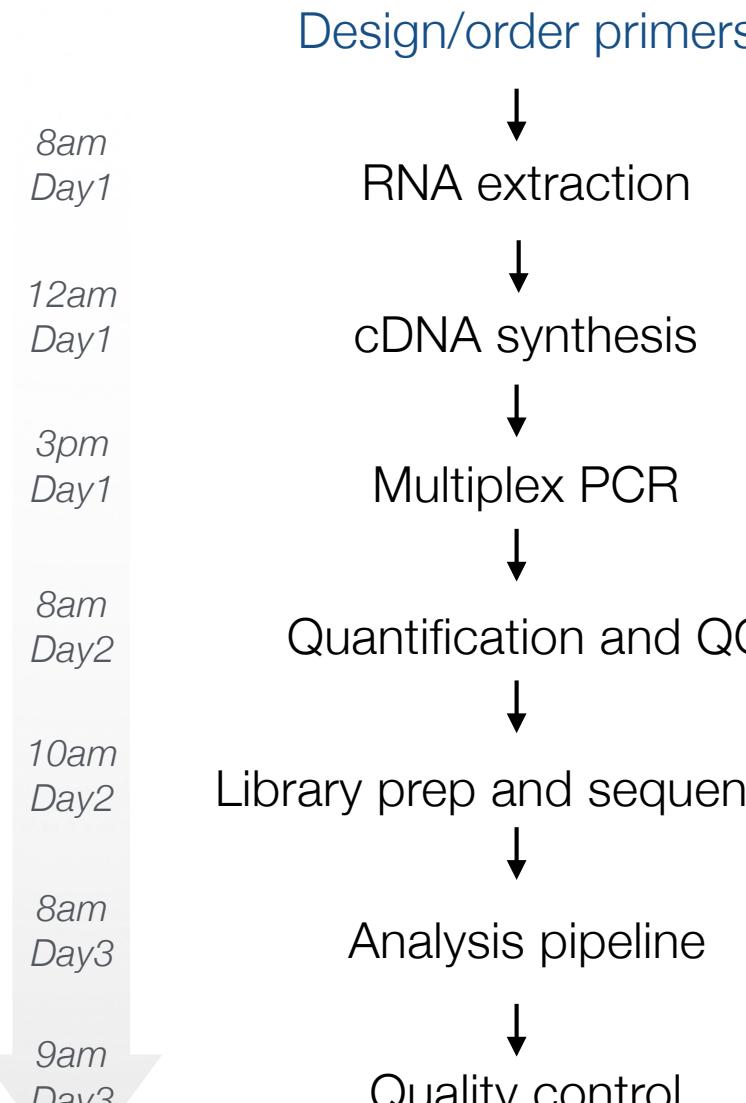
**Previous MinION work on Ebola virus:** Quick J, Loman N, et al. *Nature* 2016

**Current MinION ZiBRA protocol:** Quick J et al. *Nature Protocols* May 2017 *in press*

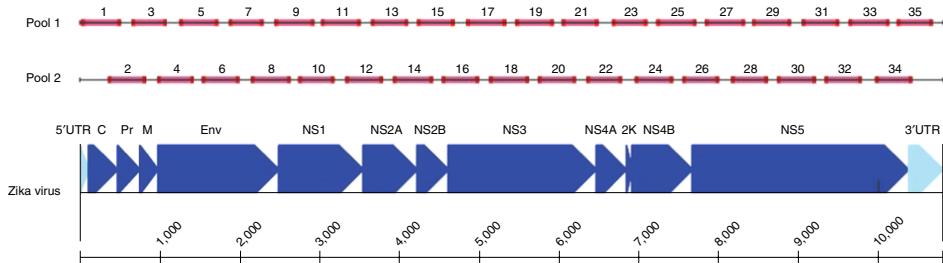
**MinION work on Zika virus in Brazil:** Faria et al. *Nature* May 2017 *in press*



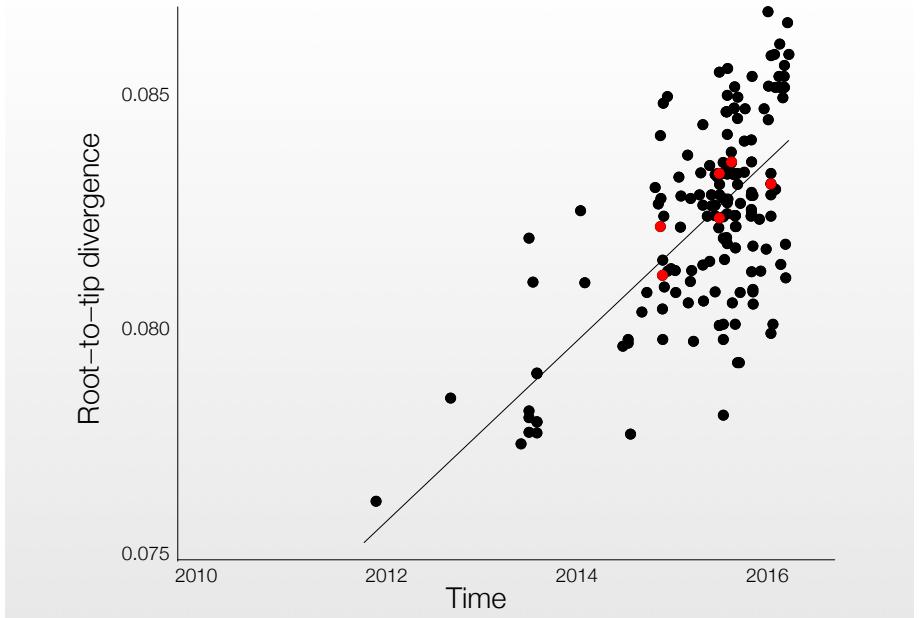
# MinION sequencing workflow for RNA virus sequencing



Web-based primer design tool:



Consensus sequence quality control:



# ZiBRA's commitment to rapid data sharing

Improving the efficiency of global outbreak control and tracking of diseases

Pre-publication data sharing, in line with WHO Consultation Data Sharing in Emergencies (1-2 Sep 2015):

*Research is essential in the context of public health emergencies. The primary purpose of such research is to advance public health, prevent illness and save lives. Researchers should always weigh the public health consequences of their actions in withholding and sharing results.<sup>1</sup>*

Twitter: <https://twitter.com/zibraproject>

Website: [zibraproject.github.io](http://zibraproject.github.io)

Github: <https://github.com/zibraproject/>

Preliminary Results: [virological.org](http://virological.org)

Virus diversity: [nextstrain.org](http://nextstrain.org)

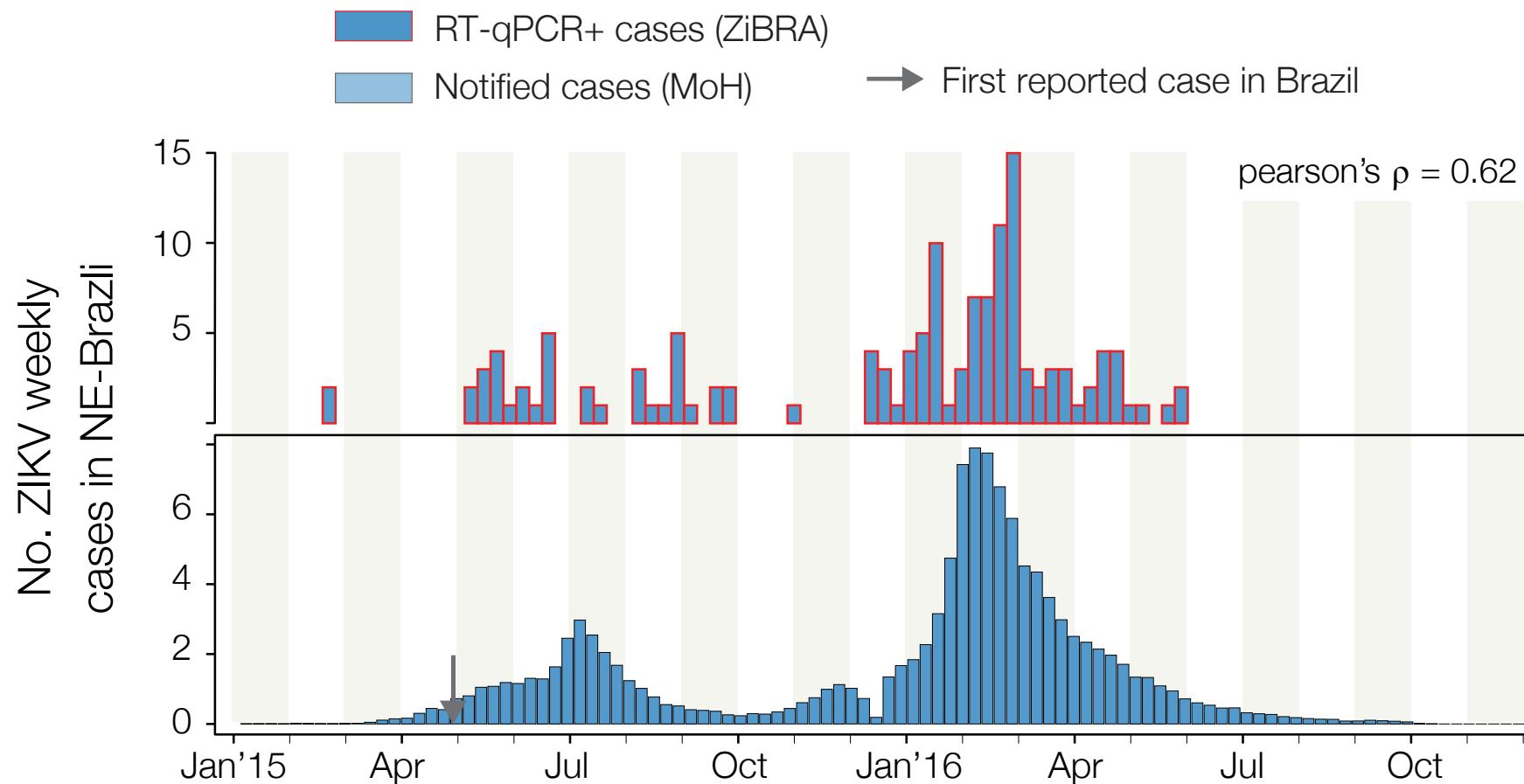
**Real-time data sharing**

<sup>1</sup>Aarestrup and Koopmans, Cell 2016

## Part 2

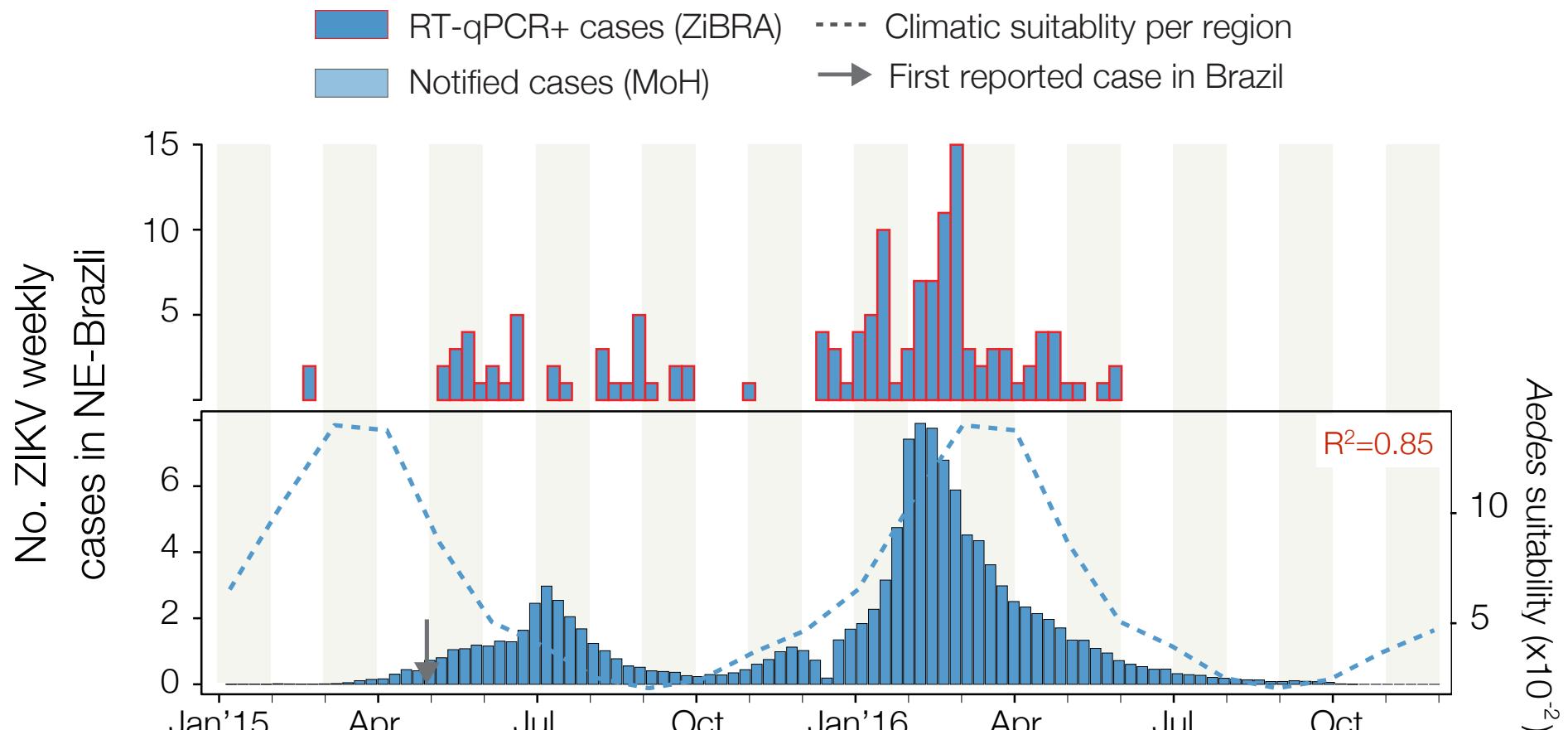
Origins and evolution of  
Zika virus in the Americas

# Zika virus MoH notified cases is positively associated with Zika RT-qPCR+ cases



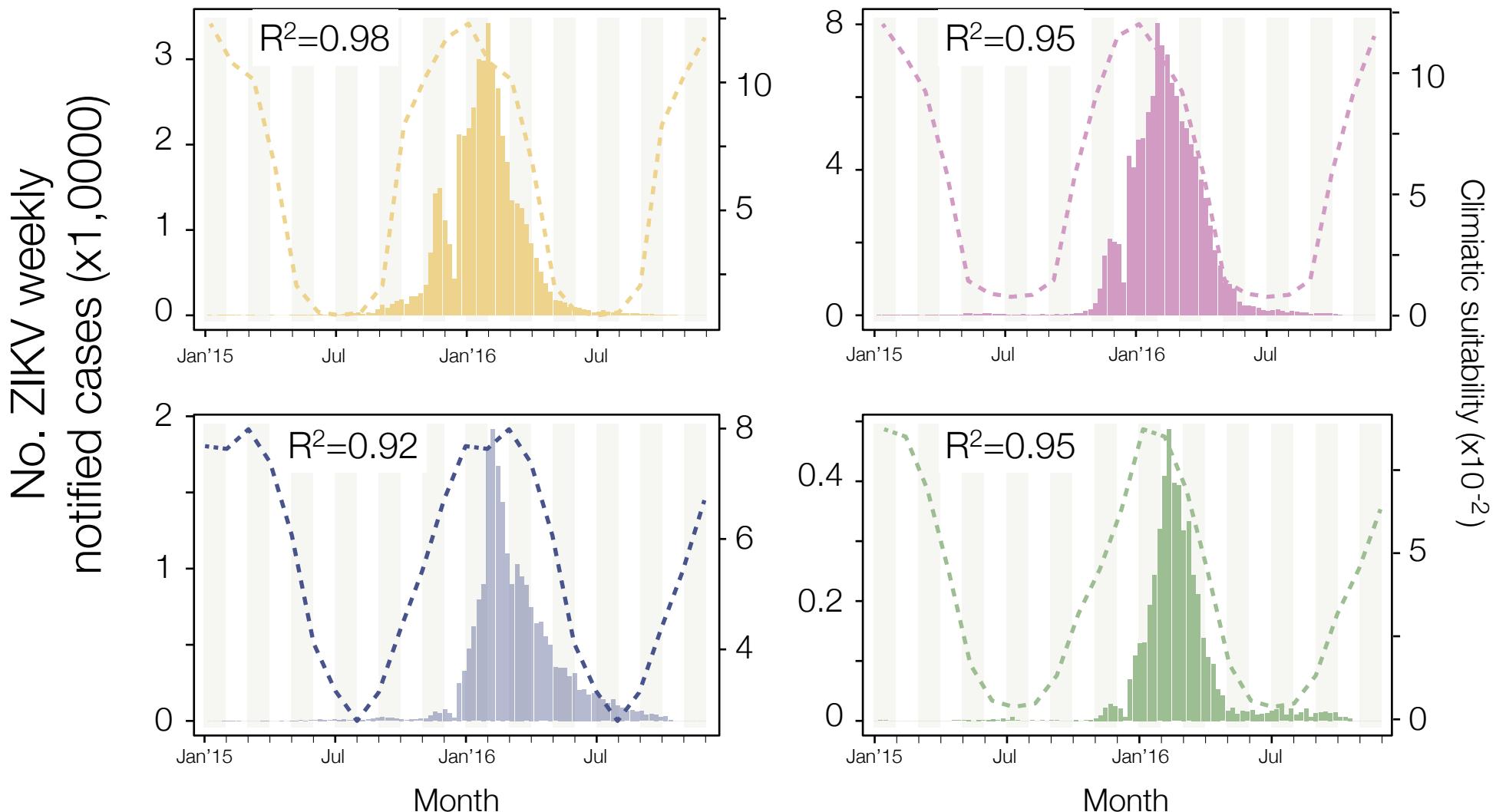
ZiBRA RT-qPCR+ data represents well the 2015-16 epidemic in NE Brazil

# Zika virus MoH notified cases is positively associated with Zika RT-qPCR+ cases **and climatic suitability**

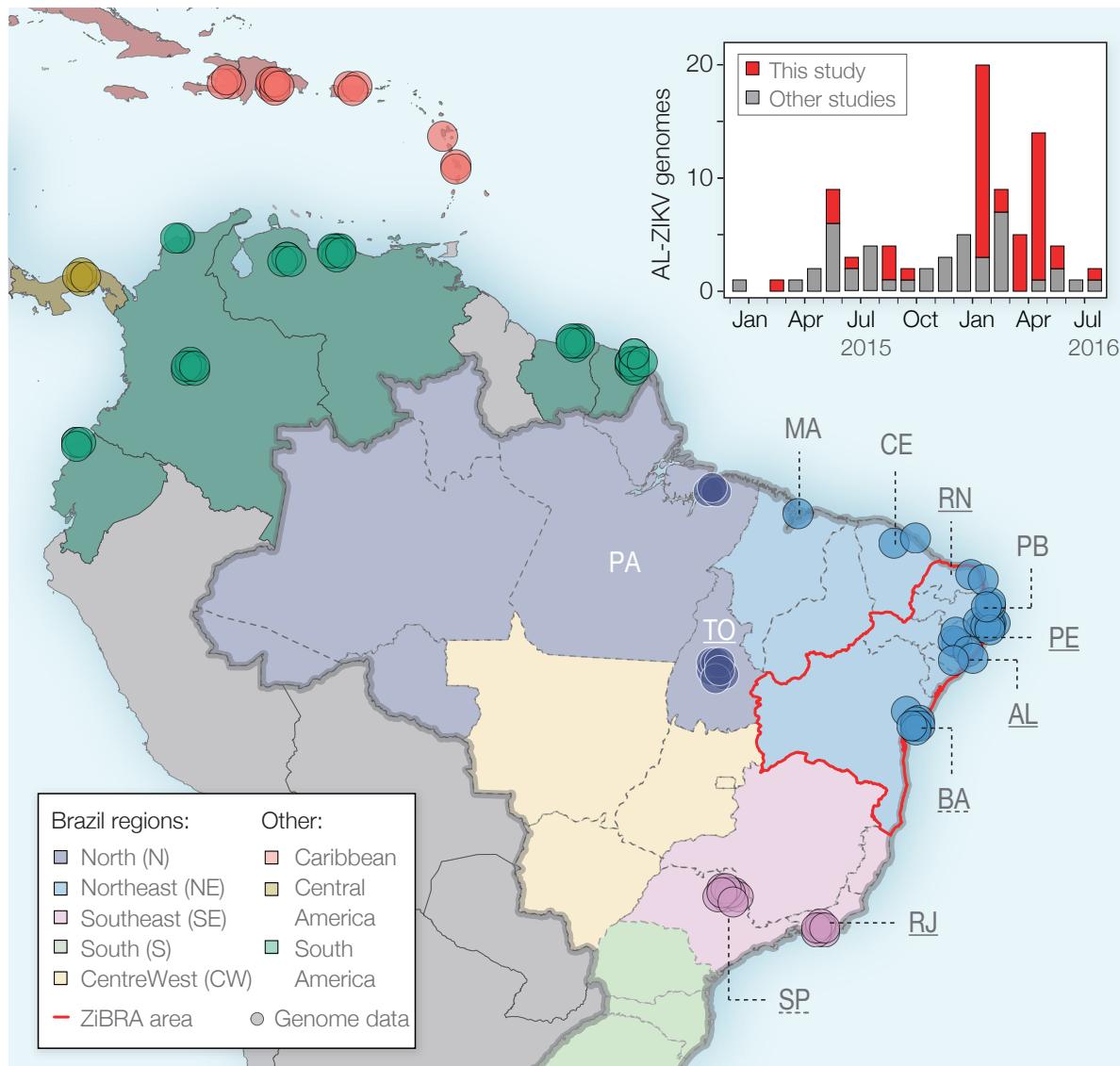


Can we predict when an epidemic will occur based on climatic factors alone?

# Regional climatic suitability strongly associated with Zika cases

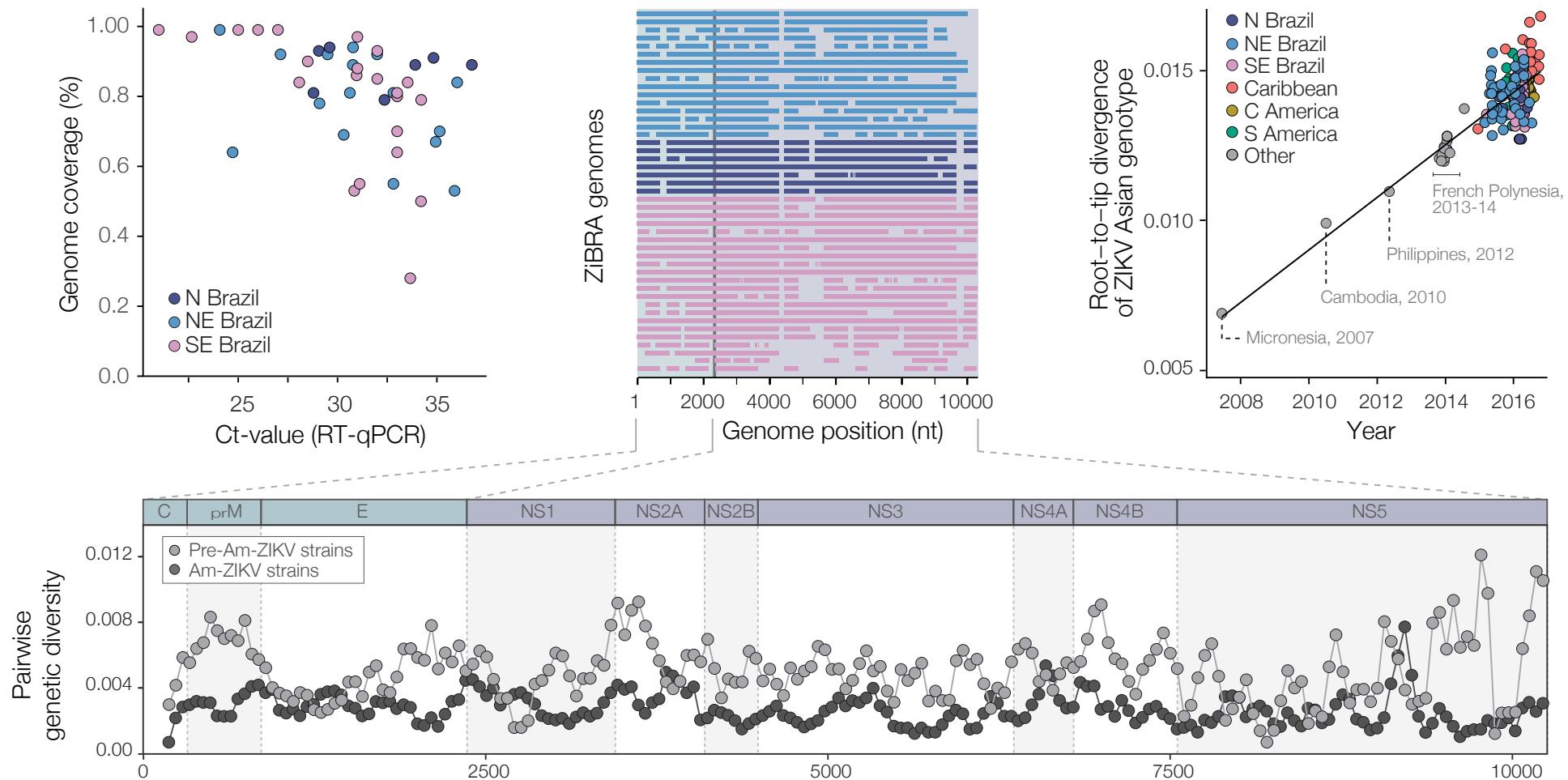


# Genome-wide analysis of ZiBRA genetic data\*



\*Nearly half (47%) of all published genomes until Feb 2017

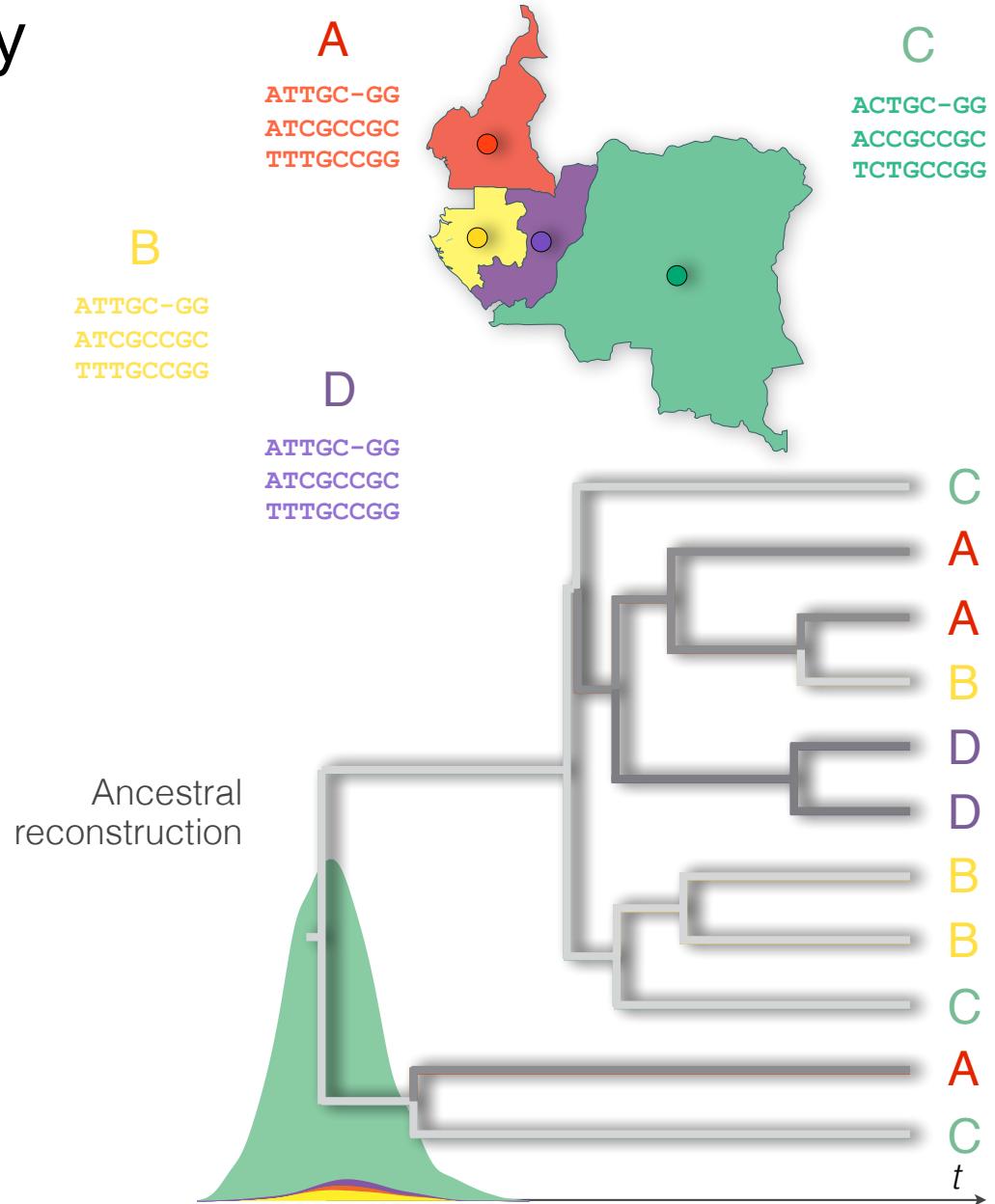
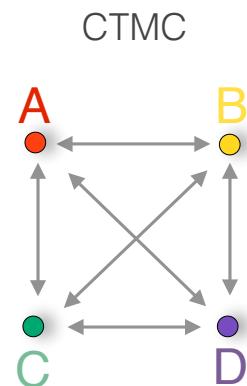
# Genomic diversity of PreAm-ZIKV and Am-ZIKV lineages



# Bayesian phylogeography

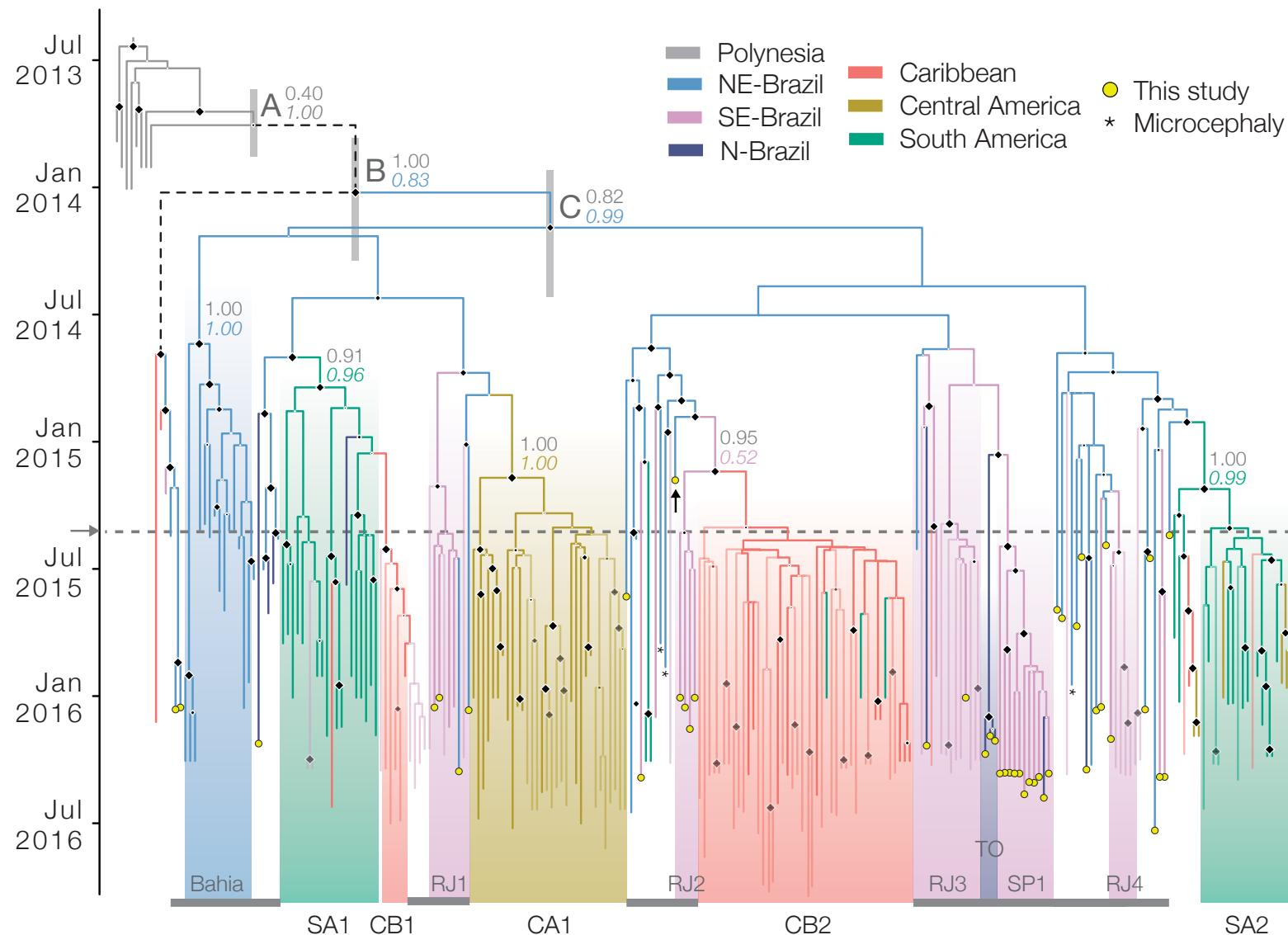
Rate matrix

$$\begin{bmatrix} A & B & C & D \\ - & \Pi_{ab} & \Pi_{ac} & \Pi_{ad} \\ \Pi_{ba} & - & \Pi_{bc} & \Pi_{bd} \\ \Pi_{ca} & \Pi_{cb} & - & \Pi_{cd} \\ \Pi_{da} & \Pi_{db} & \Pi_{dc} & - \end{bmatrix}$$



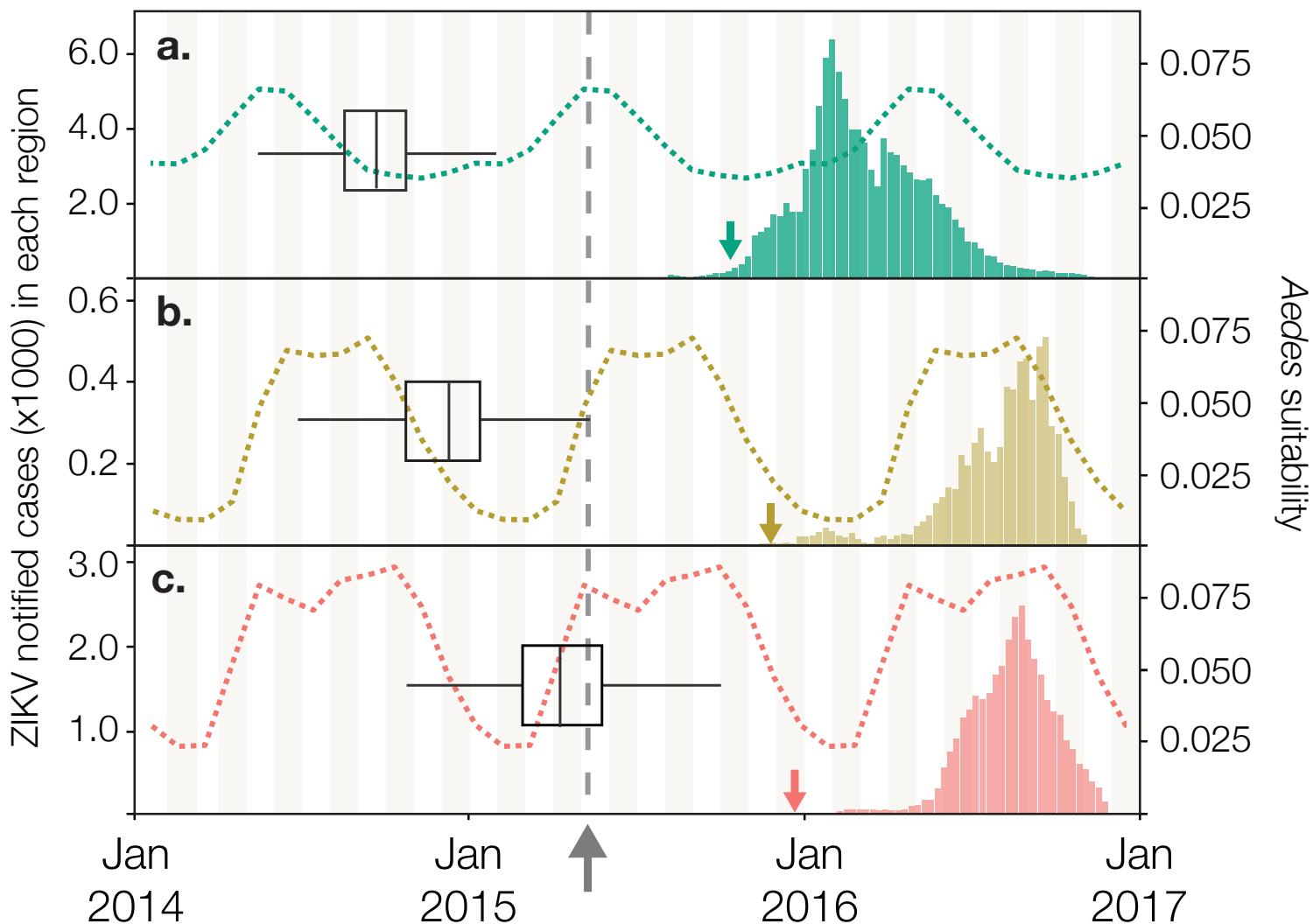
Lemey et al., *PLoS Comp Biol*, 2009; Talbi et al. *PLoS Pathog*, 2011; Drummond et al. *MBE* 2012; O'Brien et al. *MBE* 2009; Minin & Suchard, *J Math Biol* 2008 (adapted from Faria et al., *Curr Opin Virol* 2011).

# Origins of ZIKV-AL in northeast Brazil, from where it spread to southeast Brazil and beyond



Faria et al *Nature* 2017

# Cryptic circulation in several American regions for a near-complete season of transmission



Faria et al *Nature* 2017

# Phylogeographic hypothesis testing

Generalized linear model approach:

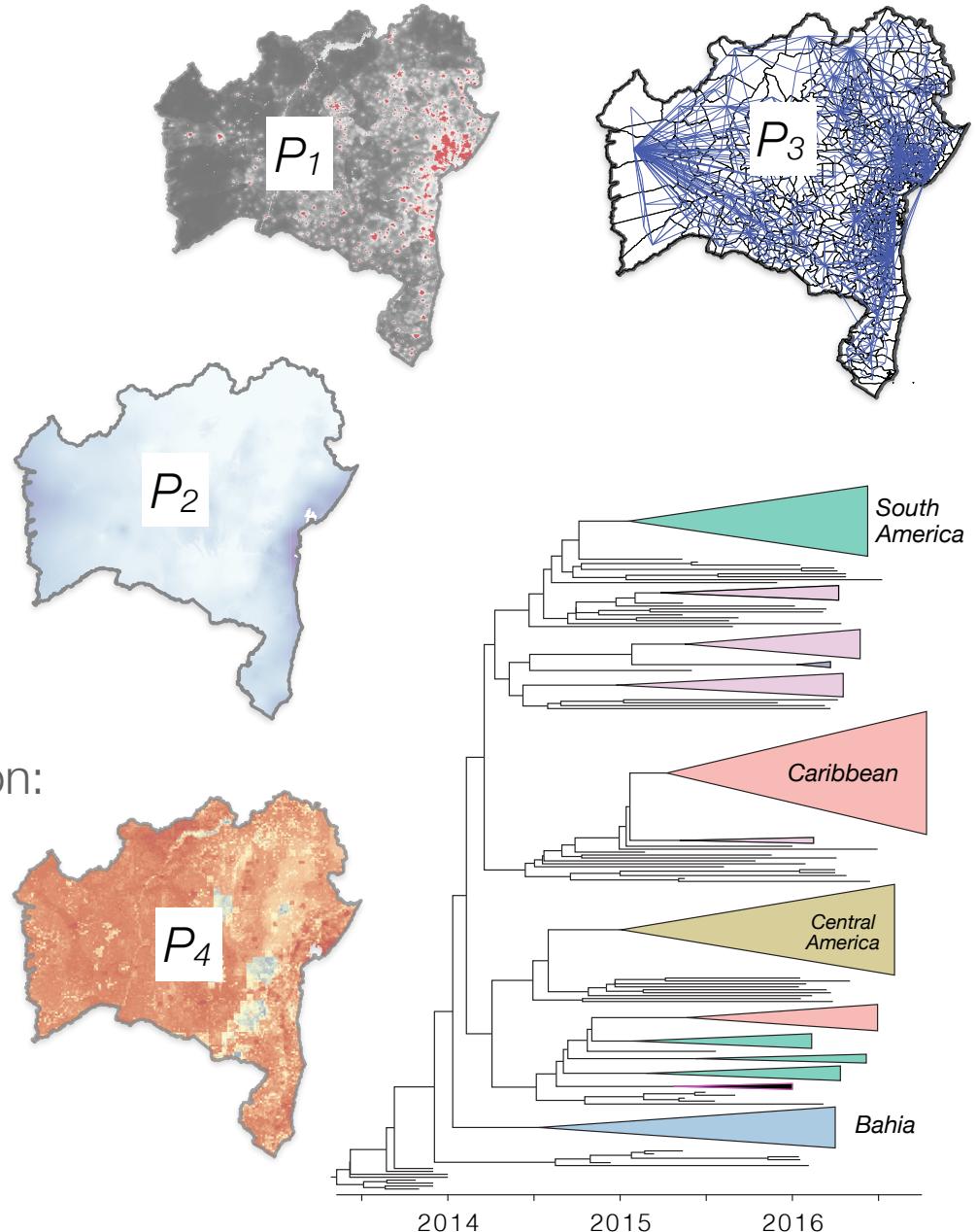
$$\begin{array}{c} \text{A} \quad \text{B} \quad \text{C} \quad \text{D} \\ \boxed{- \quad \Pi_{ab} \quad \Pi_{ac} \quad \Pi_{ad}} \\ \Pi_{ba} \quad - \quad \Pi_{bc} \quad \Pi_{bd} \\ \Pi_{ca} \quad \Pi_{cb} \quad - \quad \Pi_{cd} \\ \Pi_{da} \quad \Pi_{db} \quad \Pi_{dc} \quad - \end{array} \quad \left[ \begin{array}{c} \text{A} \\ \text{B} \\ \text{C} \\ \text{D} \end{array} \right]$$

$P_n$  as potential predictors for rate of migration:

$$\log \lambda_{AB} = \delta_{\log P_1} \beta_{\log P_1} \log P_{1AB} +$$
$$\delta_{\log P_2} \beta_{\log P_2} \log P_{2AB} +$$
$$\delta_{\log P_n} \beta_{\log P_n} \log P_{nAB}$$

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support weight predictor

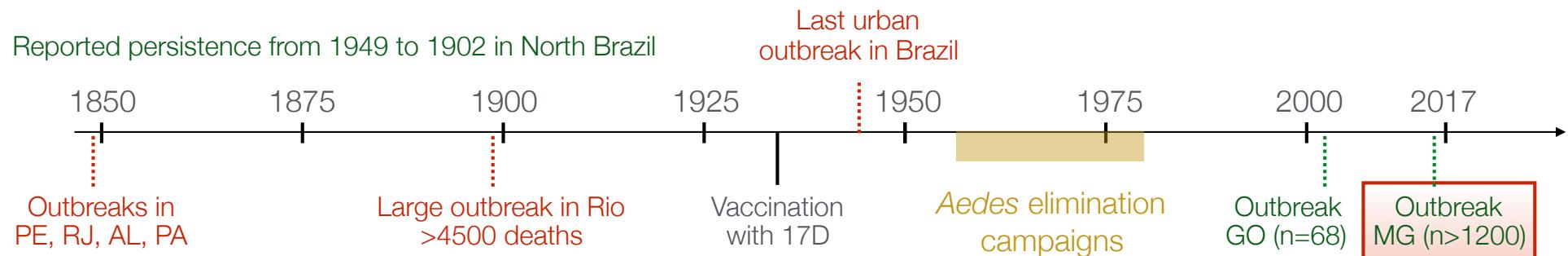


Lemey et al. *PLoS Pathogens* 2014 & Faria et al *Phil Trans R Soc B* 2013

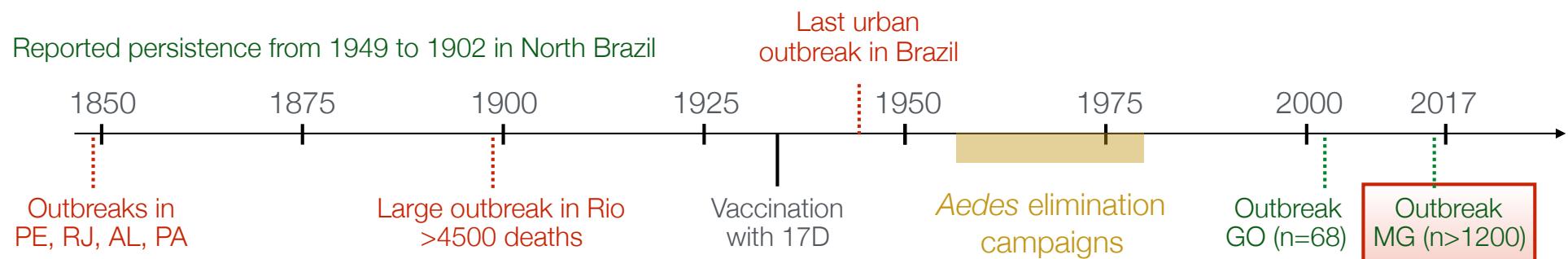
## Part 3

Real-time surveillance of the Yellow fever  
virus outbreak in Minas Gerais and beyond

# Natural history of the Yellow fever virus outbreaks in Brazil

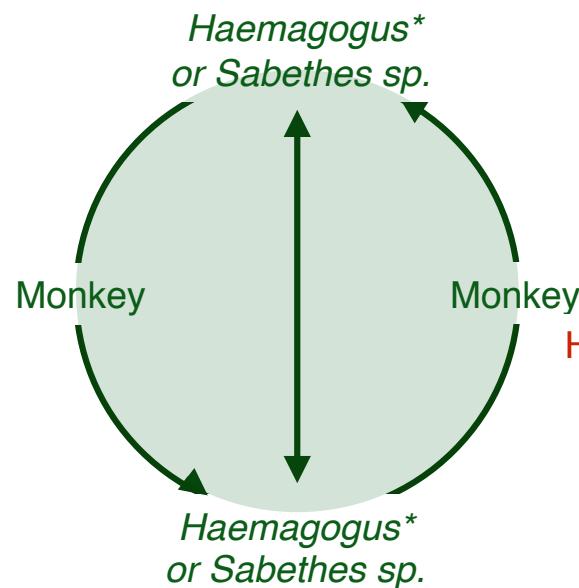


# Natural history of the Yellow fever virus outbreaks in Brazil

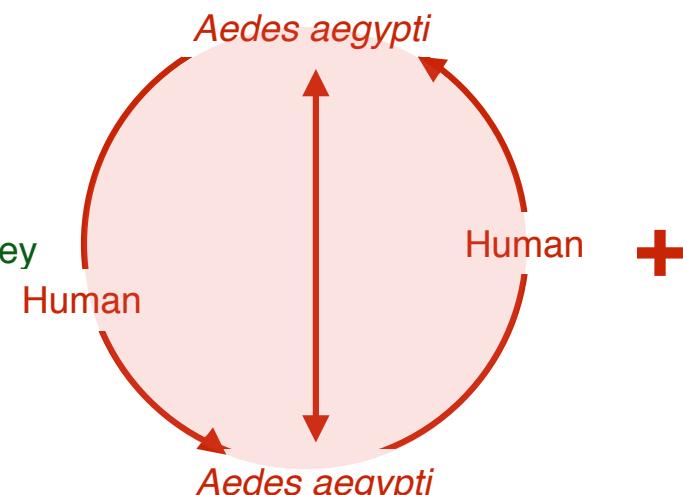


Vainio & Cutts,  
WHO 1981

## Transmission cycles in South America

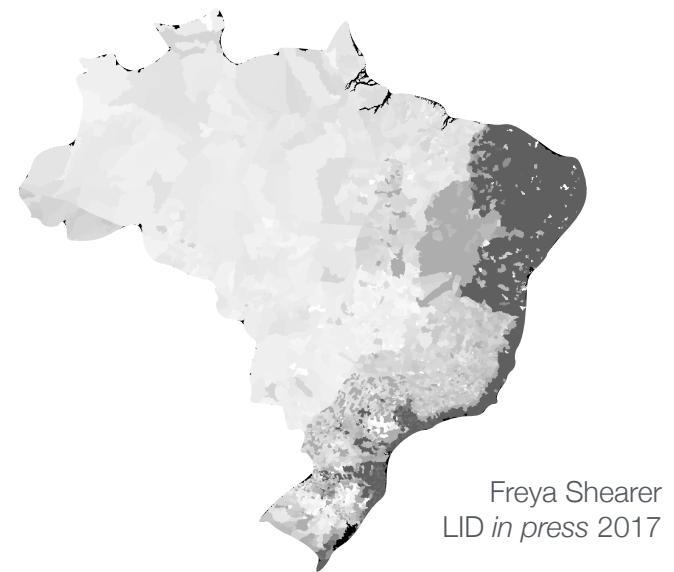


Jungle yellow fever



Urban yellow fever

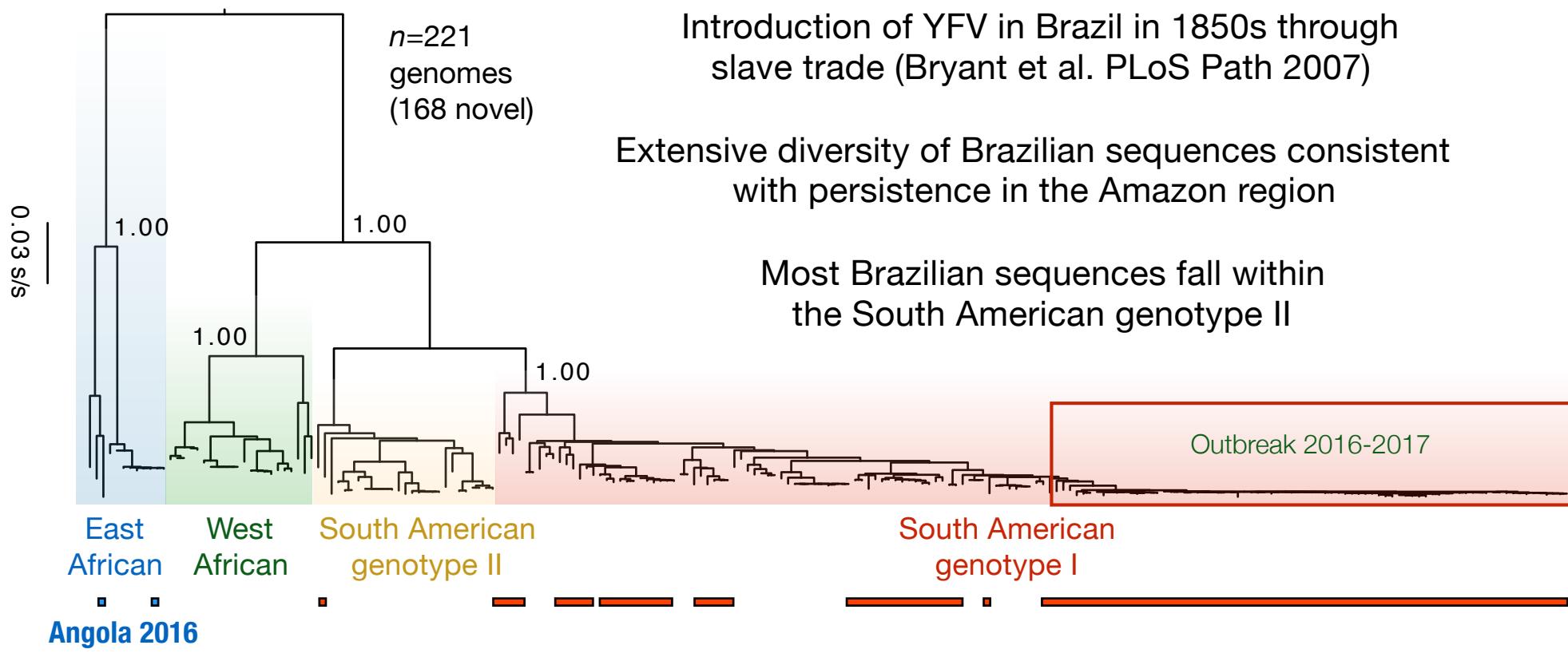
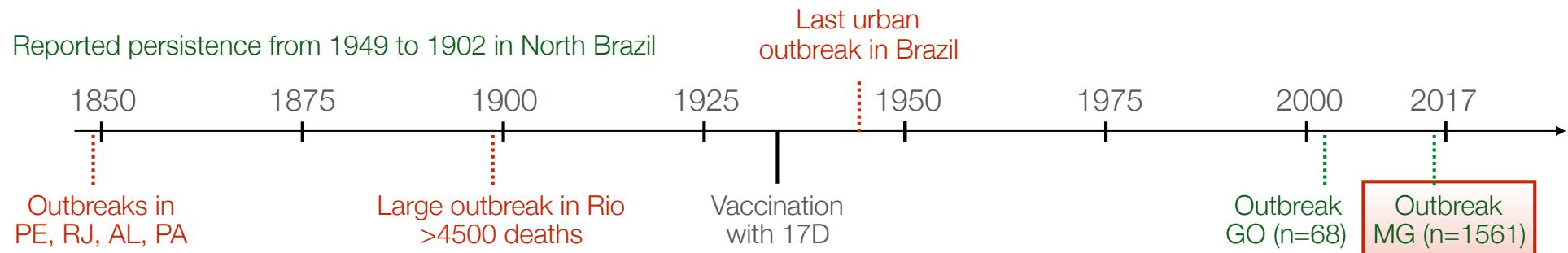
## Yellow Fever vaccination coverage Brazil 2016



Vaccination coverage:

<20 20-40 40-60 60-80 >80%

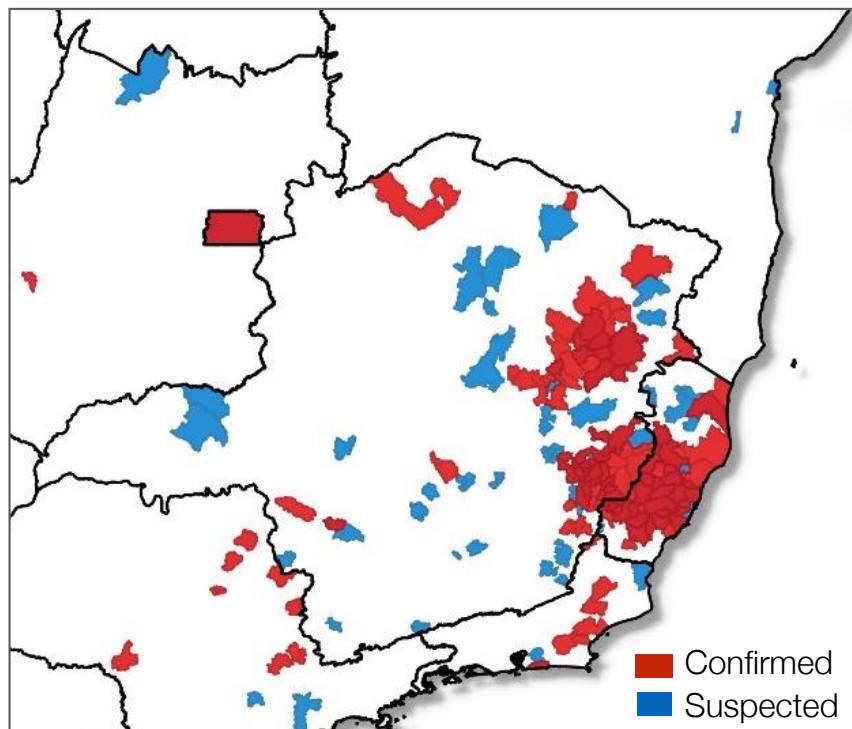
# Natural history of the Yellow fever virus outbreaks in Brazil



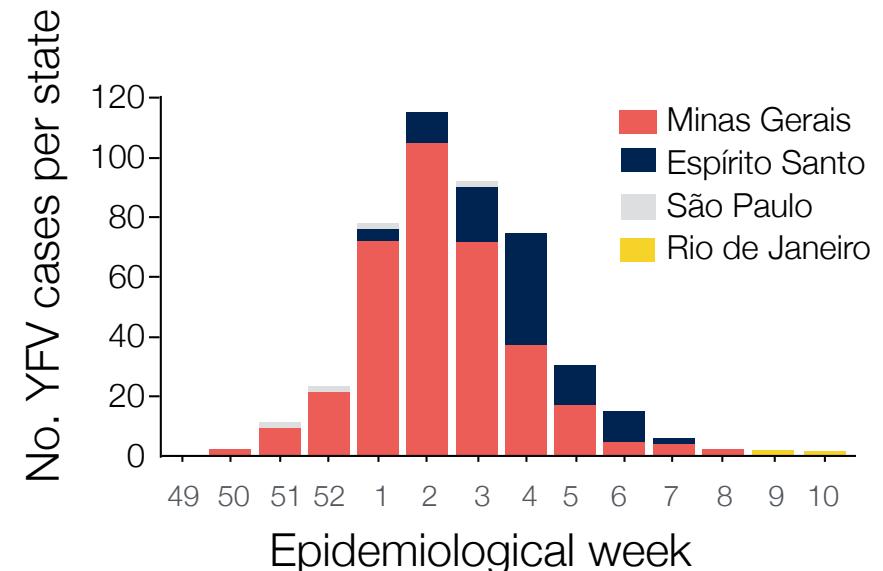
near real-time phylodynamics

# Yellow fever virus outbreak 2016-2017, southeast Brazil

Dec 2016: first confirmed cases in MG  
31% overall case-fatality rate (ECDC 2017)  
Mar 2017: 1561 cases, 264 deaths  
Sylvatic transmission (urban transmission?)



Collaboration with Oxford/FIOCRUZ/LACEN/MoH/PAHO/IEC



Genomic diversity of circulating YFV lineages?

Vaccine design & diagnostic methods

Evolutionary origins of the ongoing outbreak in Minas Gerais (MG)?

-> insight into source location and transmission cycle

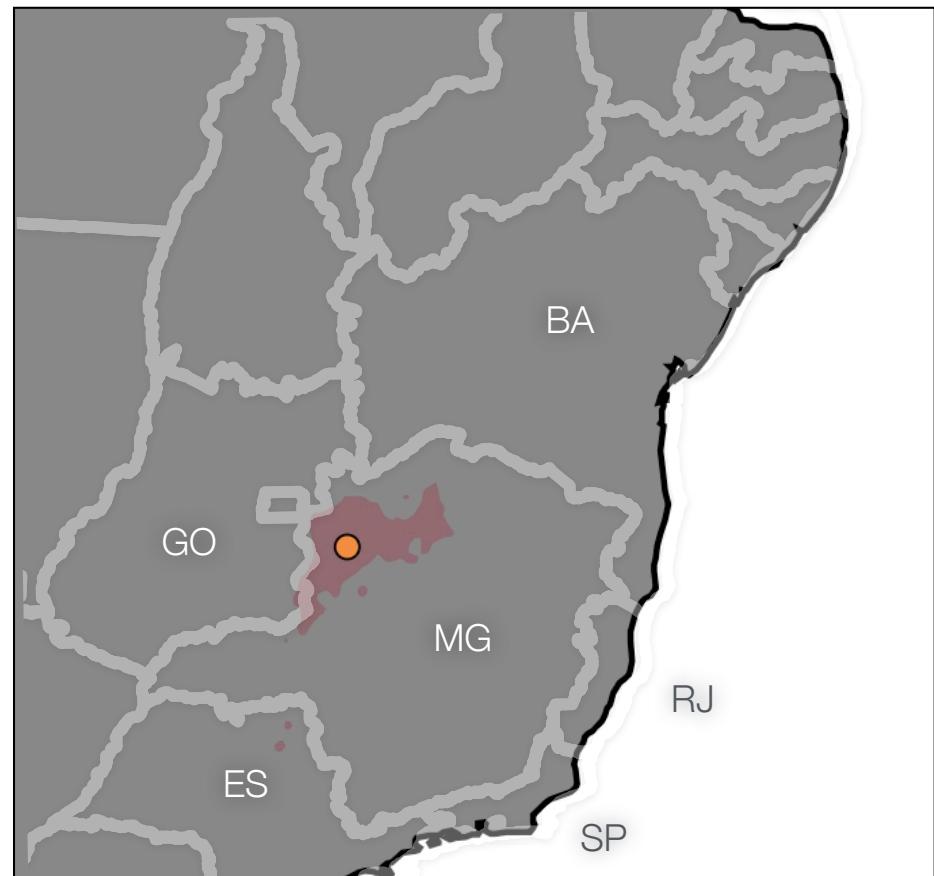
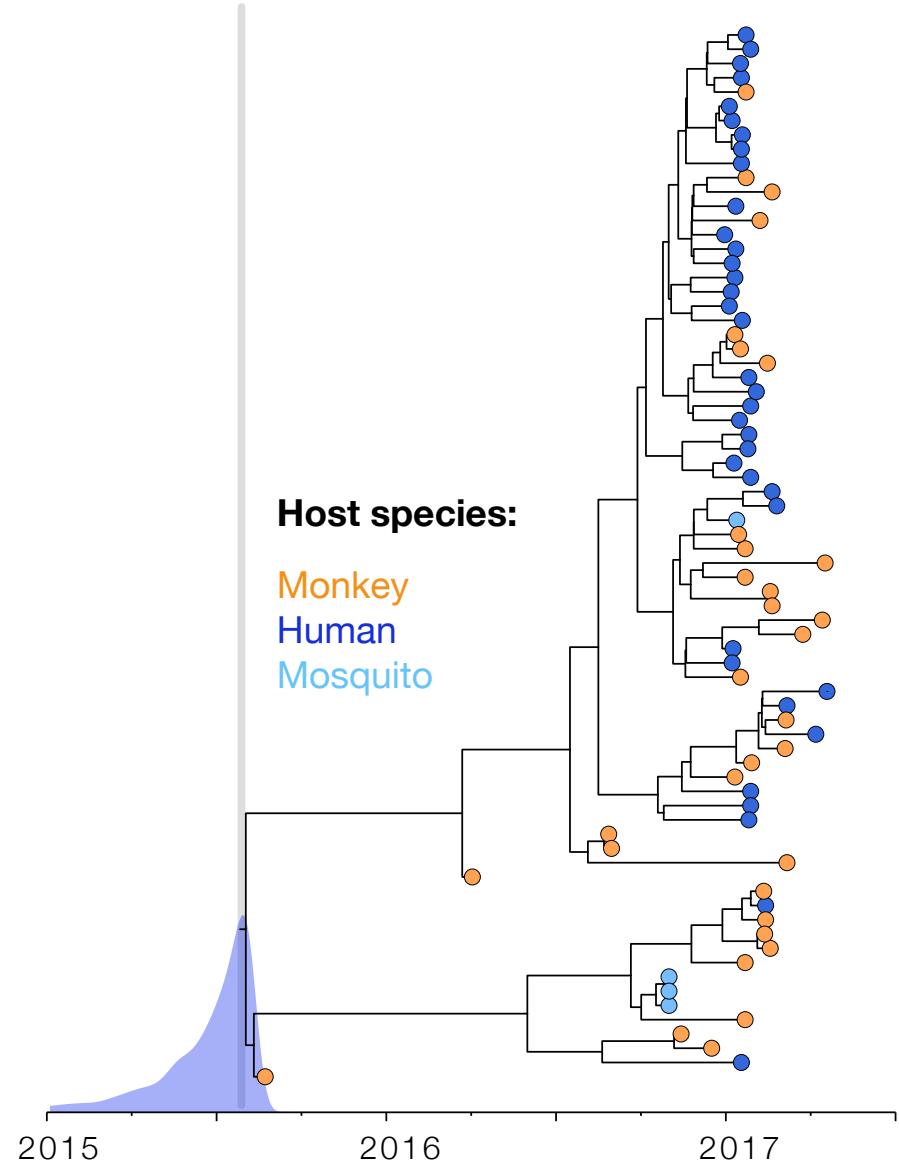
Patterns and drivers of virus spread in different MG host populations?

-> Build predictive models virus spread and forecasting

Are there associations between changes in YFV genome  
and disease severity such as convulsions or death?

No such associations have been found yet, but  
currently there are only 2 genomes available

# Wave of spread towards the Atlantic region



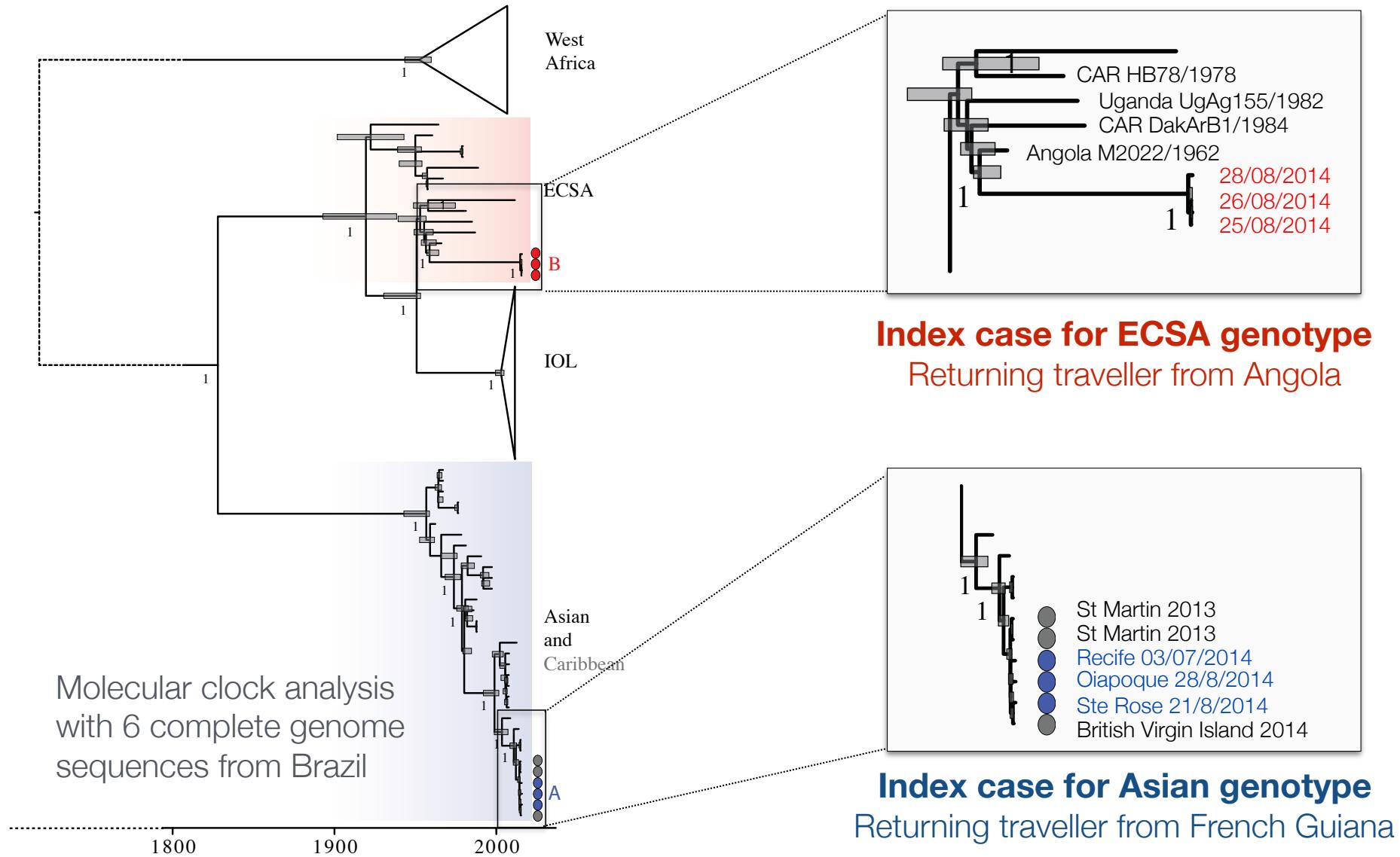
# Preliminary results on the YFV 2016-17 outbreak

- Genetic analyses suggest that the current YFV outbreak in southeast Brazil resulted most likely from a recent introduction from the north of Brazil (Amazon region) in or around mid 2015;
  - Scenario is consistent with a large YFV sylvatic outbreak aided by human mobility (27 July 2017: are the new YFV cases in Bolivia linked to MG outbreak?);
  - The virus then spread silently and rapidly in the southeast region towards the Atlantic coast (mean branch velocity: 71, 48-94 km/month) - with highly populated areas with little vaccination coverage.
- 
- What triggered the rapid spread of YFV in human populations?
  - What is the likelihood that *Ae. aegypti* will become involved in the future?
  - How can we combine genetic and mobility data to predict how it will spread?

## Part 4

Towards a global real-time genomic and  
epidemiological surveillance network

# CHIKV-ECSA introduced in Brazil from Angola

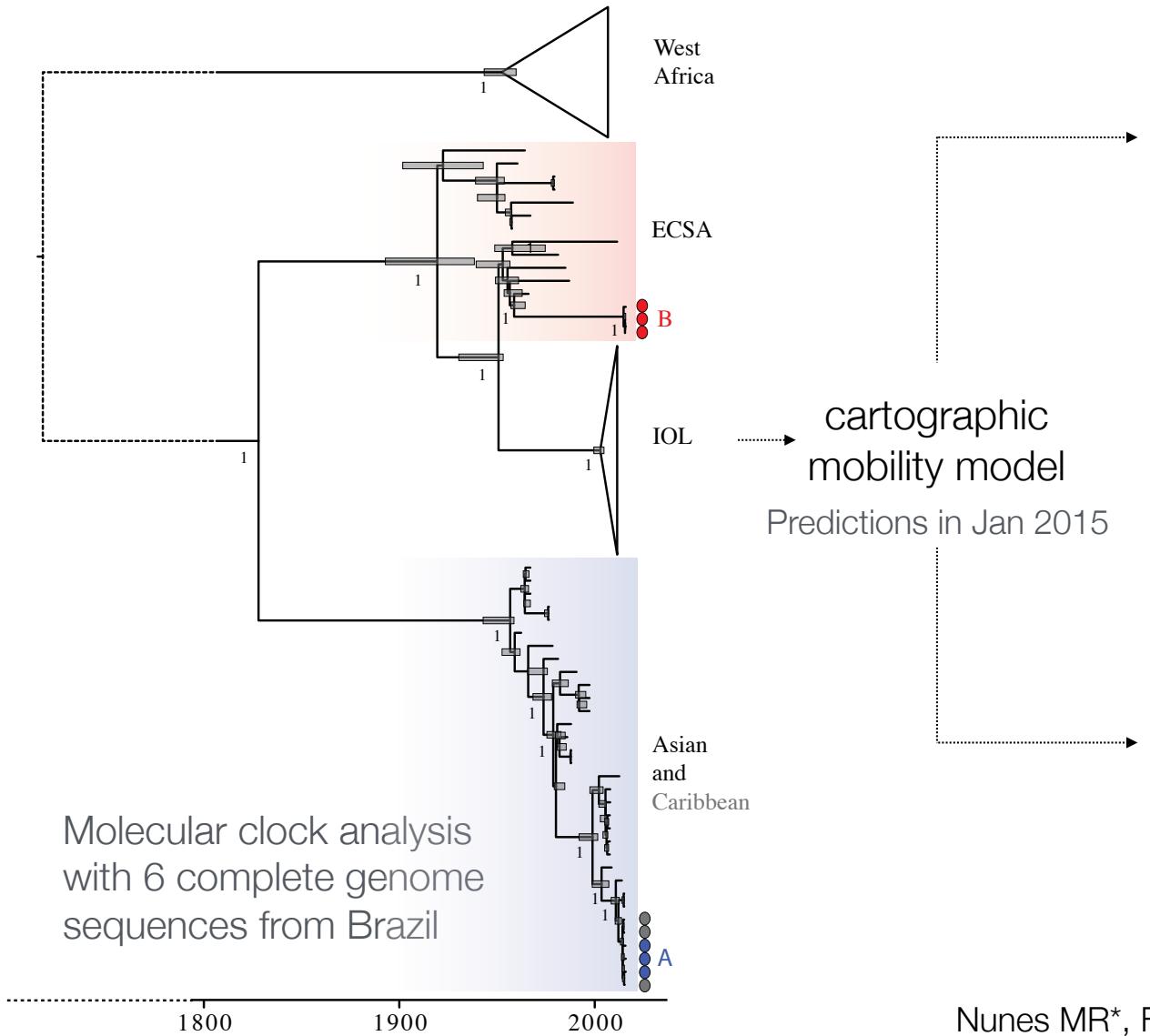


Conclusions

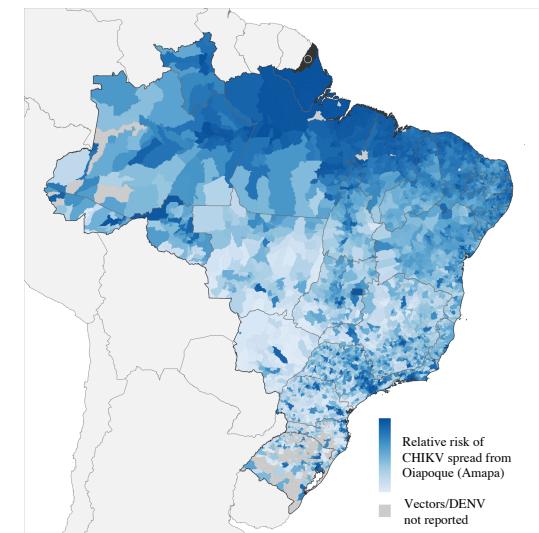
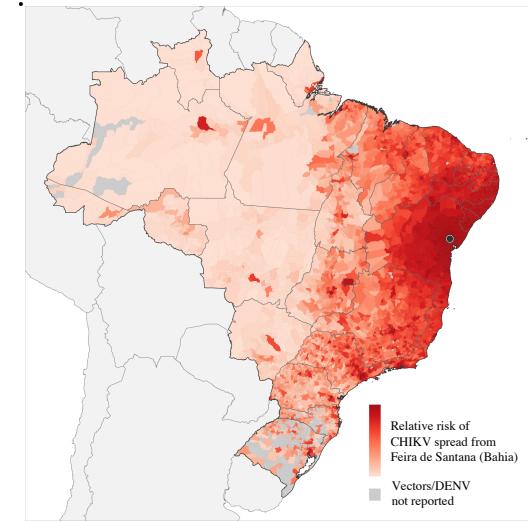
Feira de Santana, Bahia, Brazil, July 2014



# Distinct CHIKV genotypes circulating in Brazil



Nunes MR\*, Faria NR\* et al. BMC Med, April 2015



Conclusions

# Cryptic circulation of the CHIKV-ECSA in Brazil

30 Mar - 3 May 2016, 12,000 visits in 2 hospitals in Maceió, Alagoas, Brazil

70% visits w/ symptoms compatible with DENV, ZIKV or CHIKV

76% CHIKV+, 24% ZIKV+ (RT-qPCR)

13% CHIKV+ and ZIKV+ (RT-qPCR)

Fever (87%), arthralgia (70%), headache (44%), exanthema (30%) and myalgia (26%)

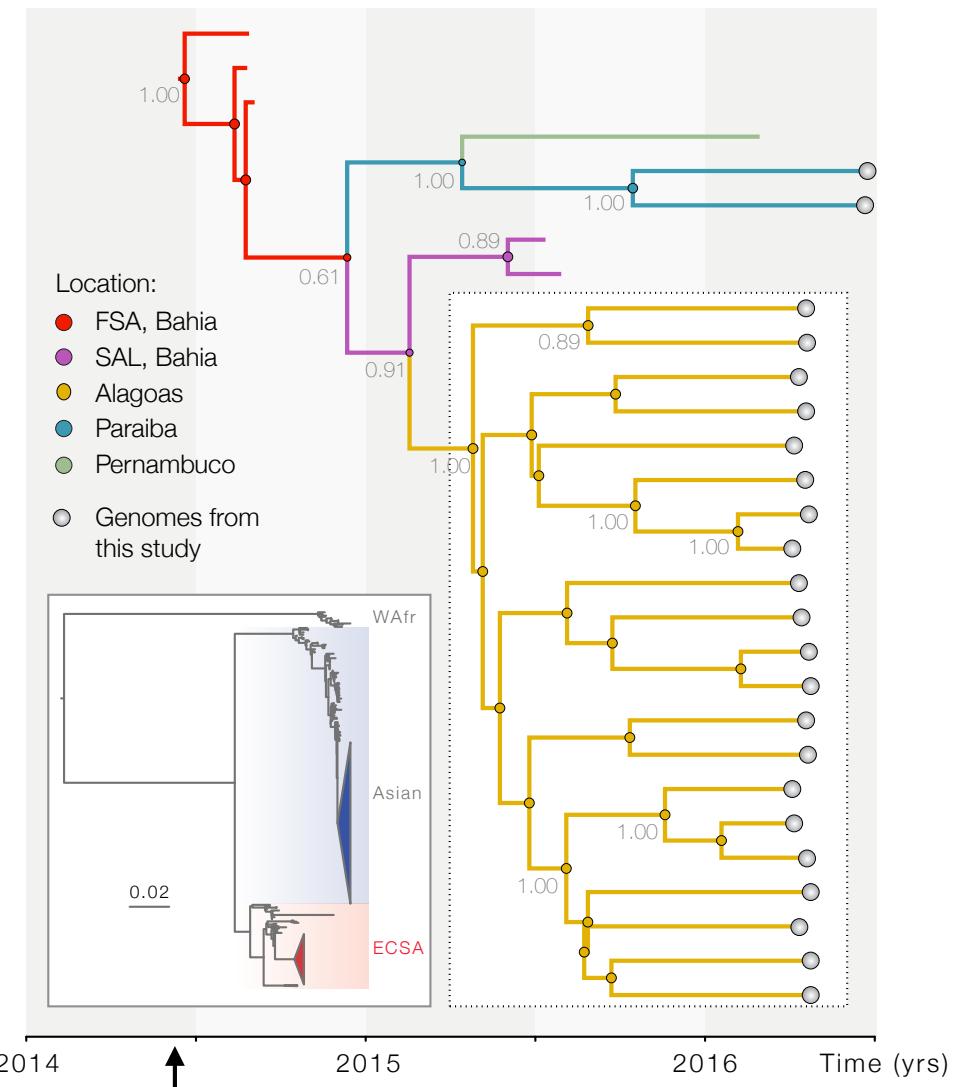
Two genotypes spreading in the Americas

Risk of CHIKV sylvatic cycle in the region

CHIKV-ECSA genotype: adaptive mutations associated with the *Ae. albopictus*?

Distribution genotypes? Cross-protection?

da Costa et al. EID, 2017 *in press*



Conclusions

# Challenges for real-time phylogenetics

Integration of multiple data sources in real-time will require new, faster models that allow analysing large amounts of data in real-time

- Most state-of-the-art models are time-prohibitive - downsampling?
- LSD (Gascuel et al. *Sys Biol* 2016), TimeTree (Neher et al *BioRxiv* 2017)

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Faster response to disease outbreak is needed to prevent future epidemics

- Real-time digital pathogen surveillance - the time is now (Gardy, Loman and Rambaut, *Genome Biology* 2015)
- Generation of outbreak genome data is now faster (36h from sample collection) and cheaper (50\$/genome) (Quick et al *Nature Protoc* 2017)

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Building a real-time *global* genomic surveillance network

- Ecology and mobility play key roles in the global spread of infectious diseases
- Data sharing for PHEIC policies (Aarestrup and Koopmans, *Cell* 2016)
- Roots, not parachutes (Yozwiak, Sabeti, Andersen, *Cell* 2016)

# Acknowledgments

Julien Thézé, Dep. Zoology, University of Oxford, UK  
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Centro de Pesquisas Gonçalo Moniz



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**Thank you**

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