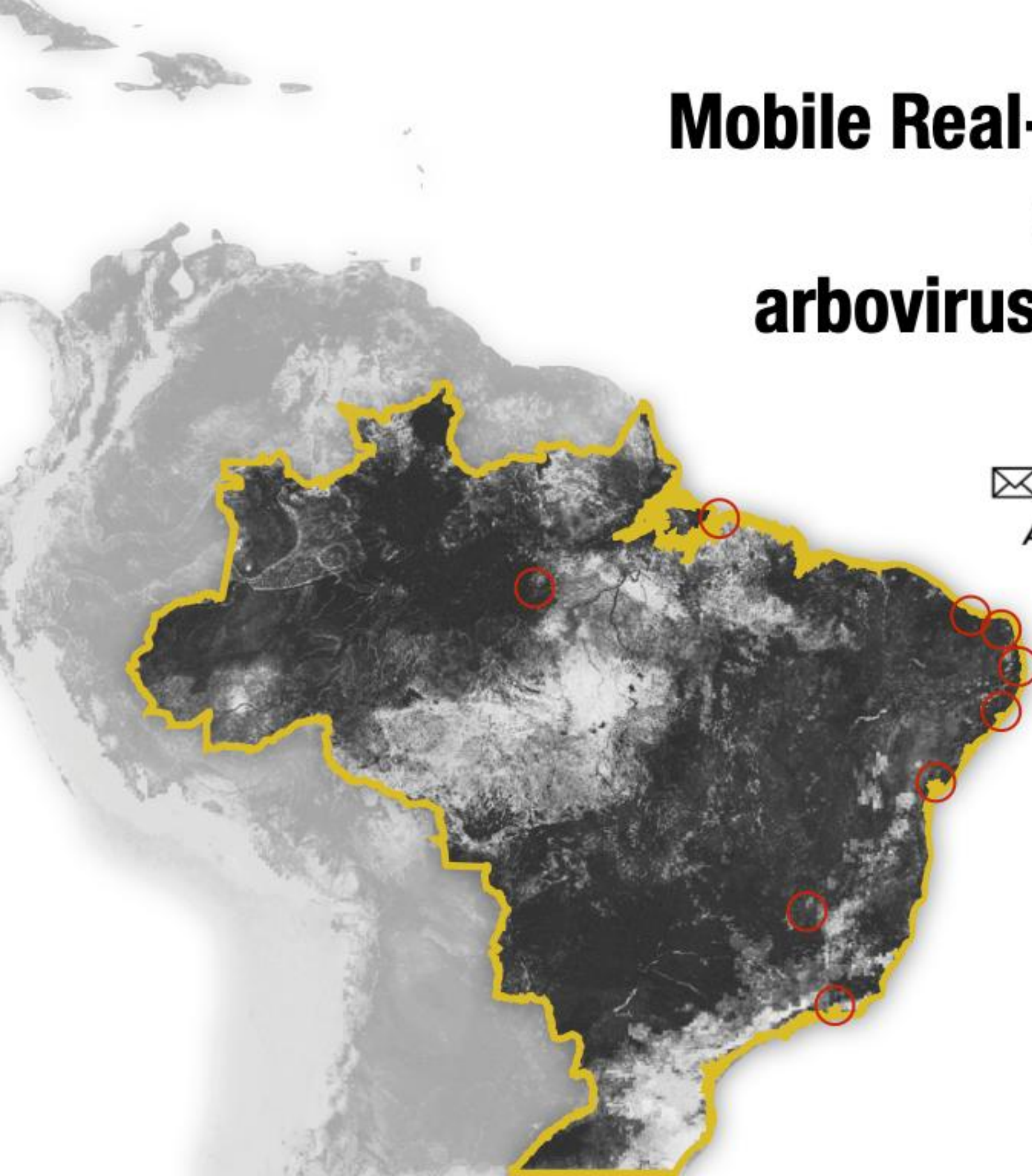


Mobile Real-time genomic of arboviruses in Brazil

Marta Giovanetti

✉ giovanetti.marta@gmail.com

ALAG – MENDONZA (AR) 2019



A person wearing a blue lab coat is working in a laboratory setting. In the foreground, a large roll of white mesh material is partially unrolled, showing some yellowish stains. The background is slightly blurred, showing laboratory equipment and shelves.

From Zika to ZIBRA

Zika in Brazil real-time analysis

Sequenciamento por nanoporos: A revolução genômica

Ian Goodfellow in Sierra Leone 2015

**Ion Torrent**
@iontorrent

 **Following**

Dr. Goodfellow .@igoodfel uses #NGS to study the Ebola Virus and its #evolution
oak.ctx.ly/r/40u6h



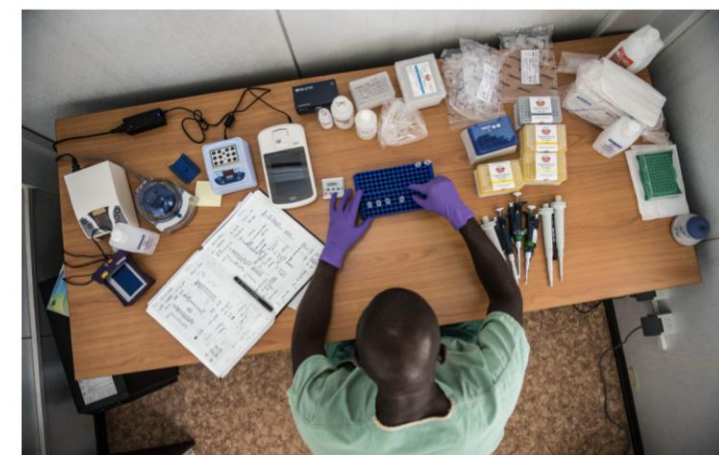
RETWEETS
13

LIKES
13



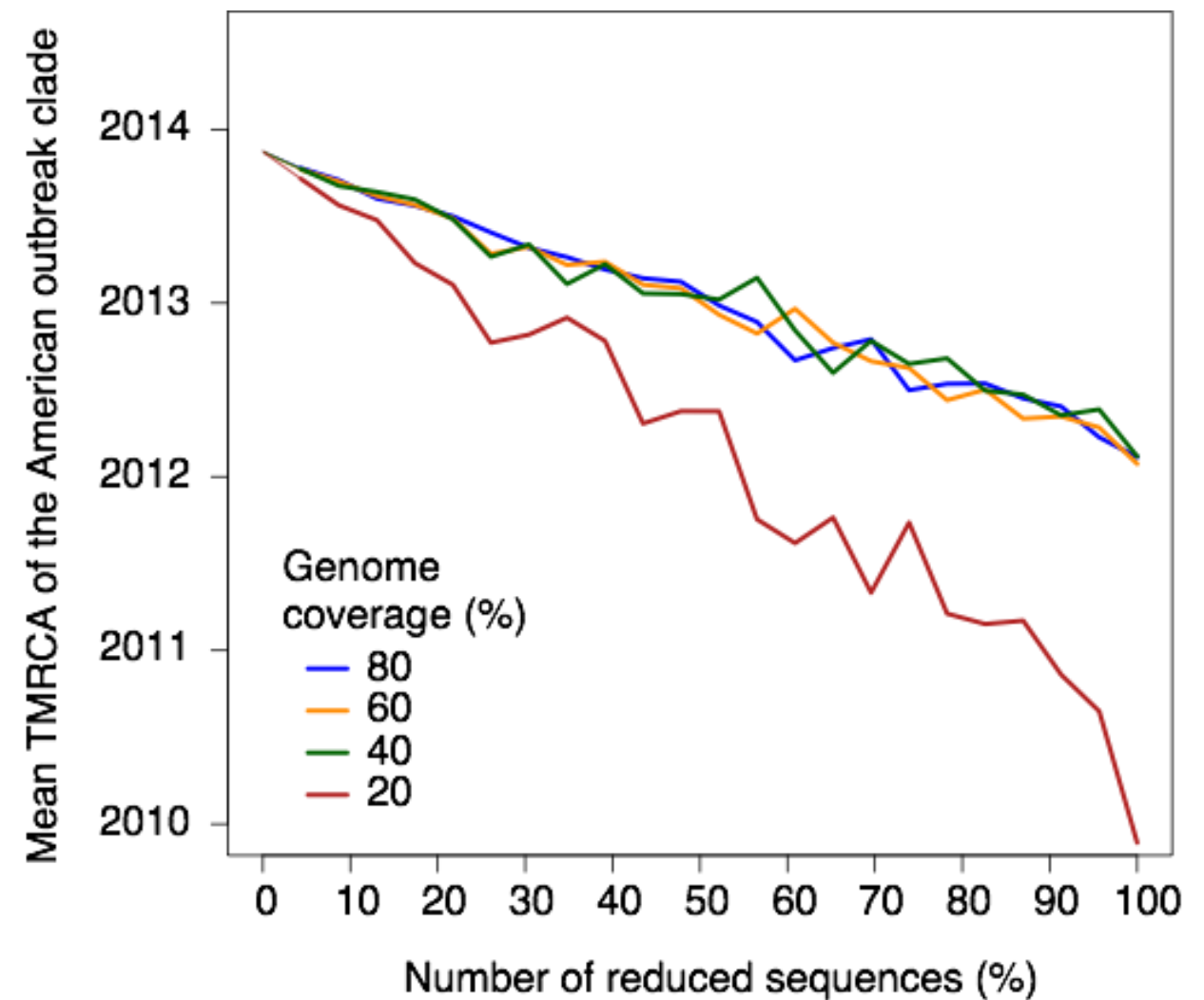
12:08 PM - 24 Nov 2015

Josh Quick in Guinea 2015

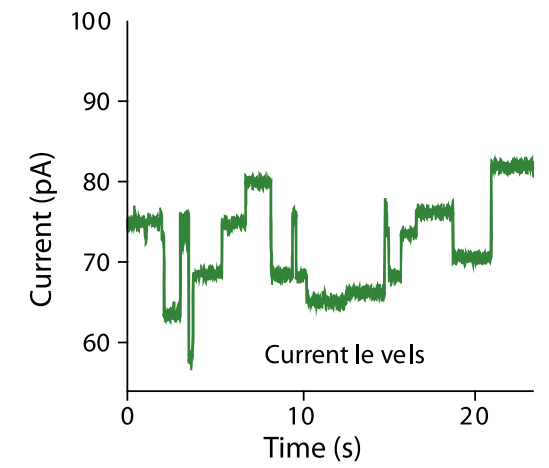
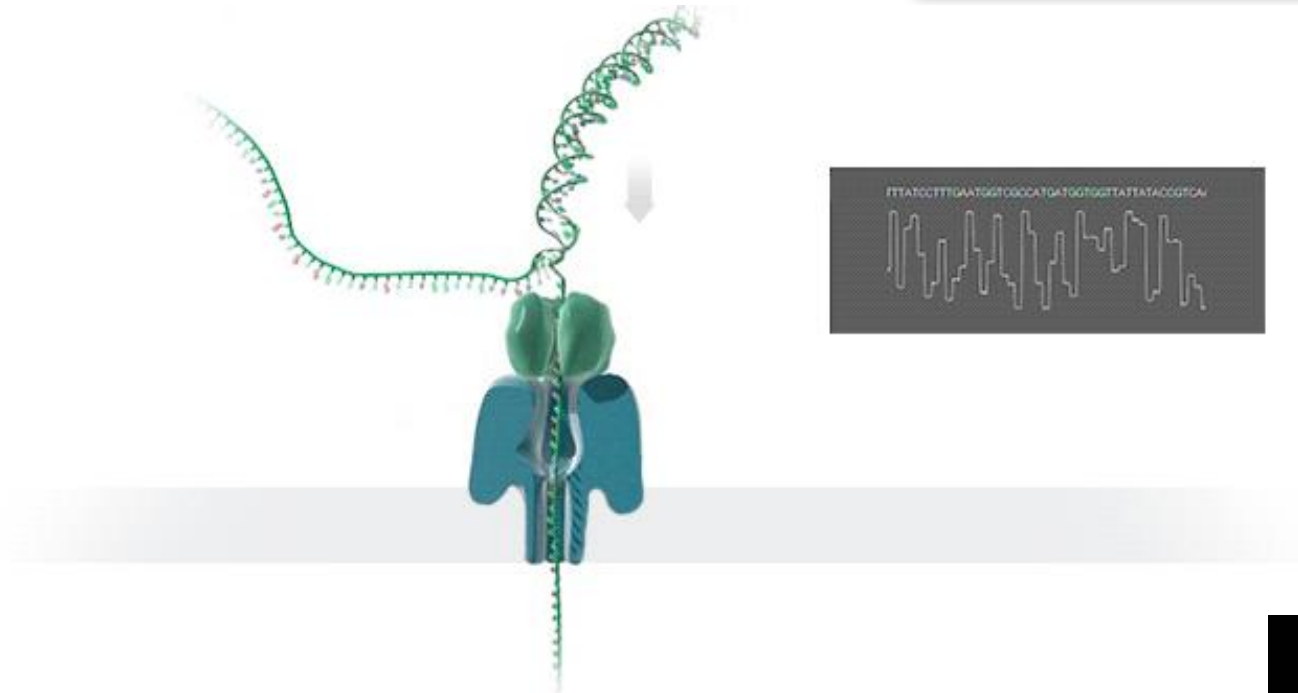
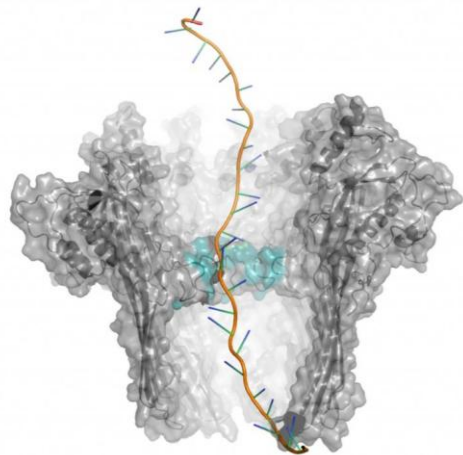
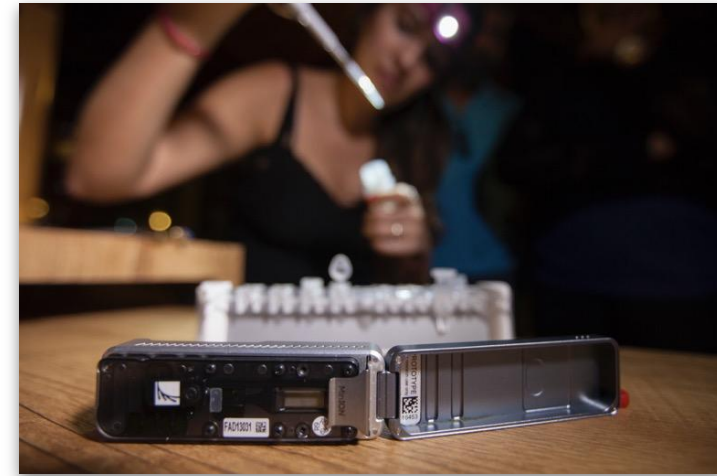


Genomas completos contêm mais informação em comparação com sequências parciais

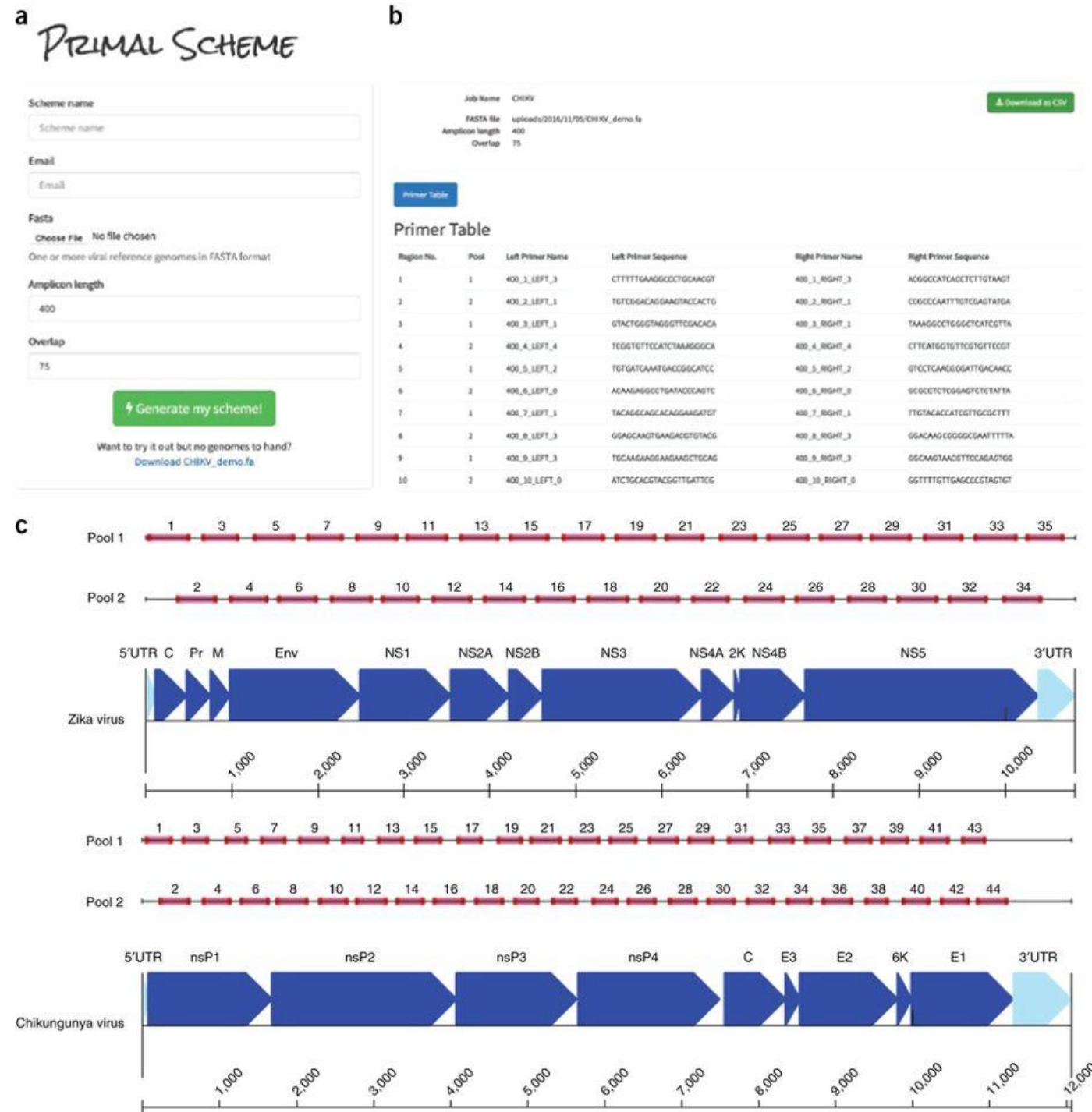
- Identificação de mutações associadas à transmissão ou gravidade da doença
- O aumento da resolução filogenética é obtido quando:
 - ▶ Sequências completas são geradas em vez de sequências parciais
 - ▶ O intervalo de amostragem entre coleções é mais amplo (nem sempre possível)



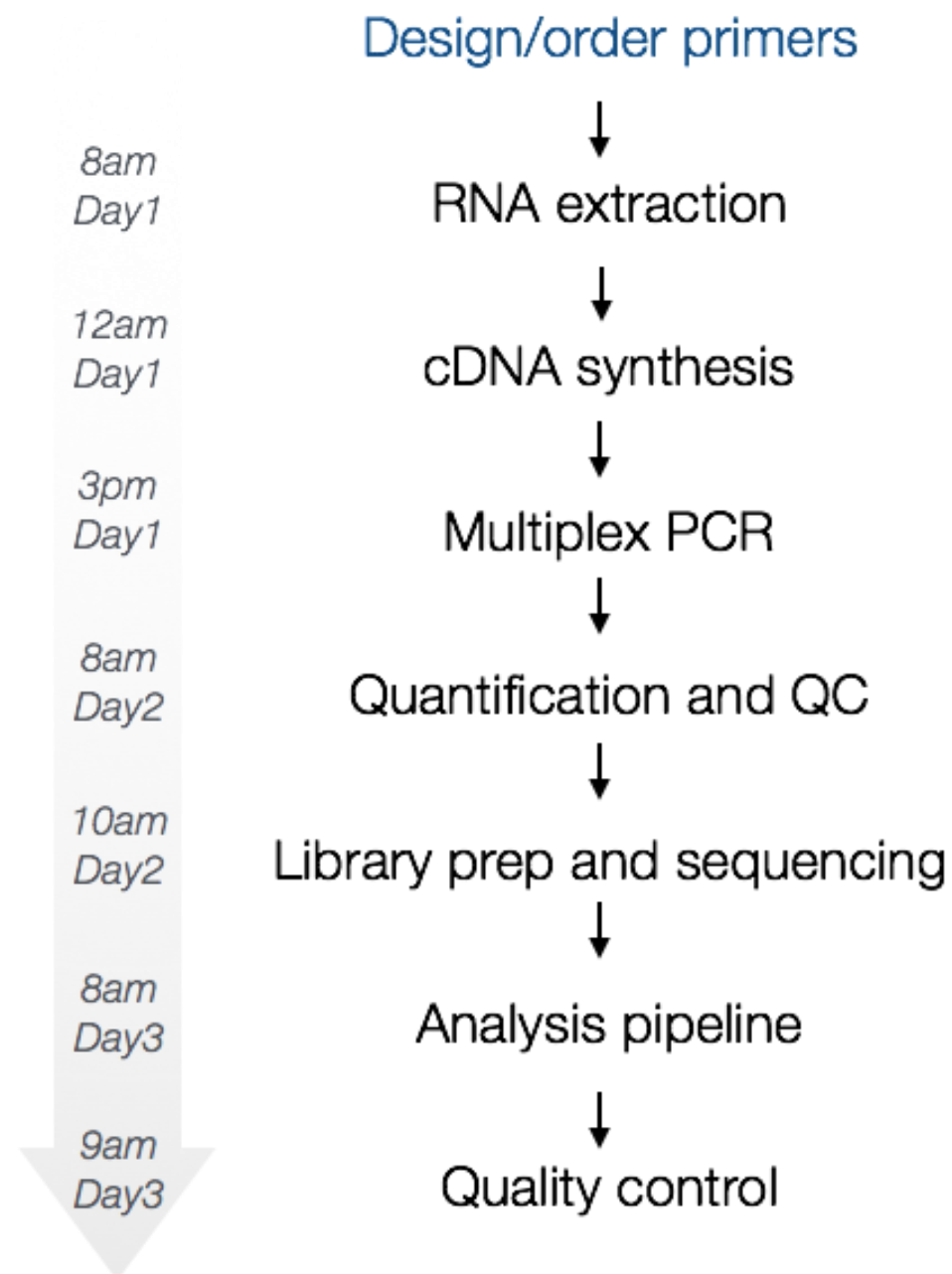
Projeto ZiBRA



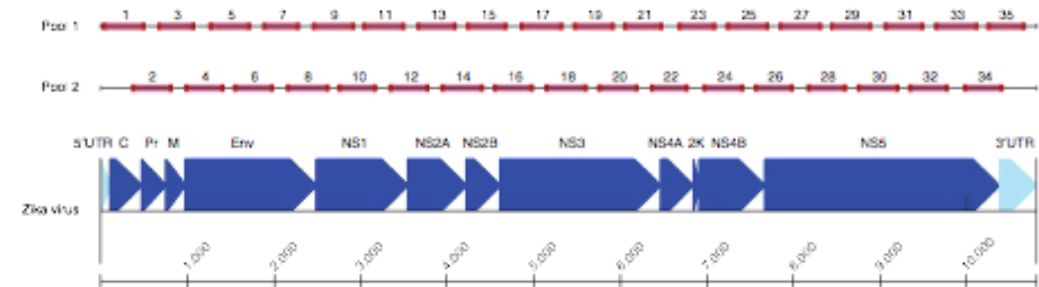
Projeto ZiBRA



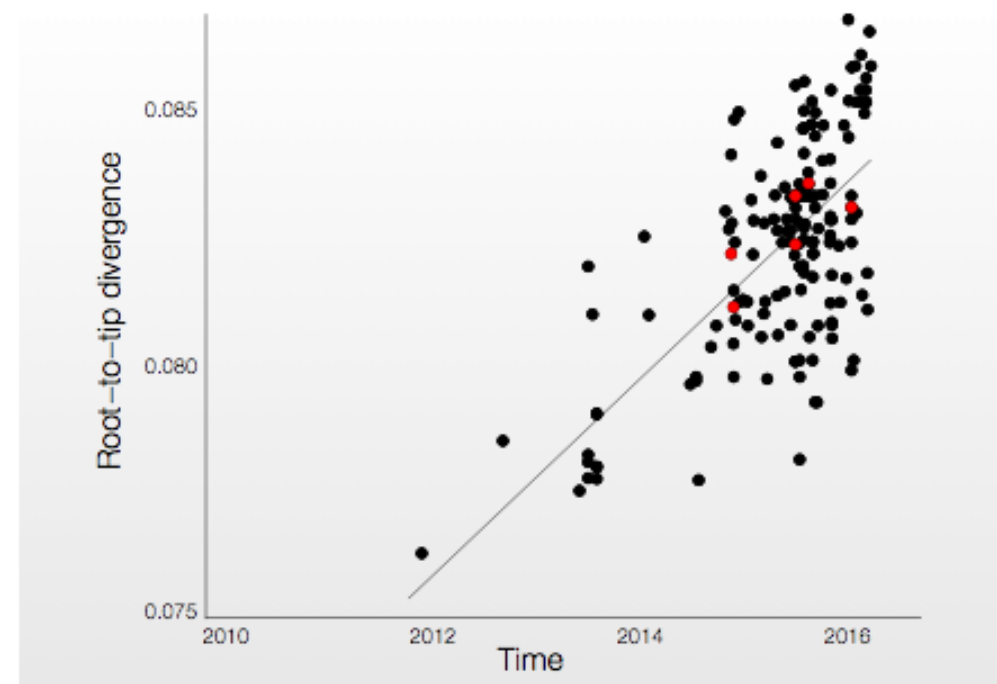
MinION sequencing workflow for RNA viruses



Web-based primer design tool:



Consensus sequence quality control:



Quick et al. Nature Protocols 2017

Parte 1

Vigilância Genômica do vírus ZIKV

Parte 2

Vigilância Genômica do vírus CHIKV

Parte 3

Vigilância Genômica do vírus YFV

Parte 1

Vigilância Genômica do vírus ZIKV

Parte 2

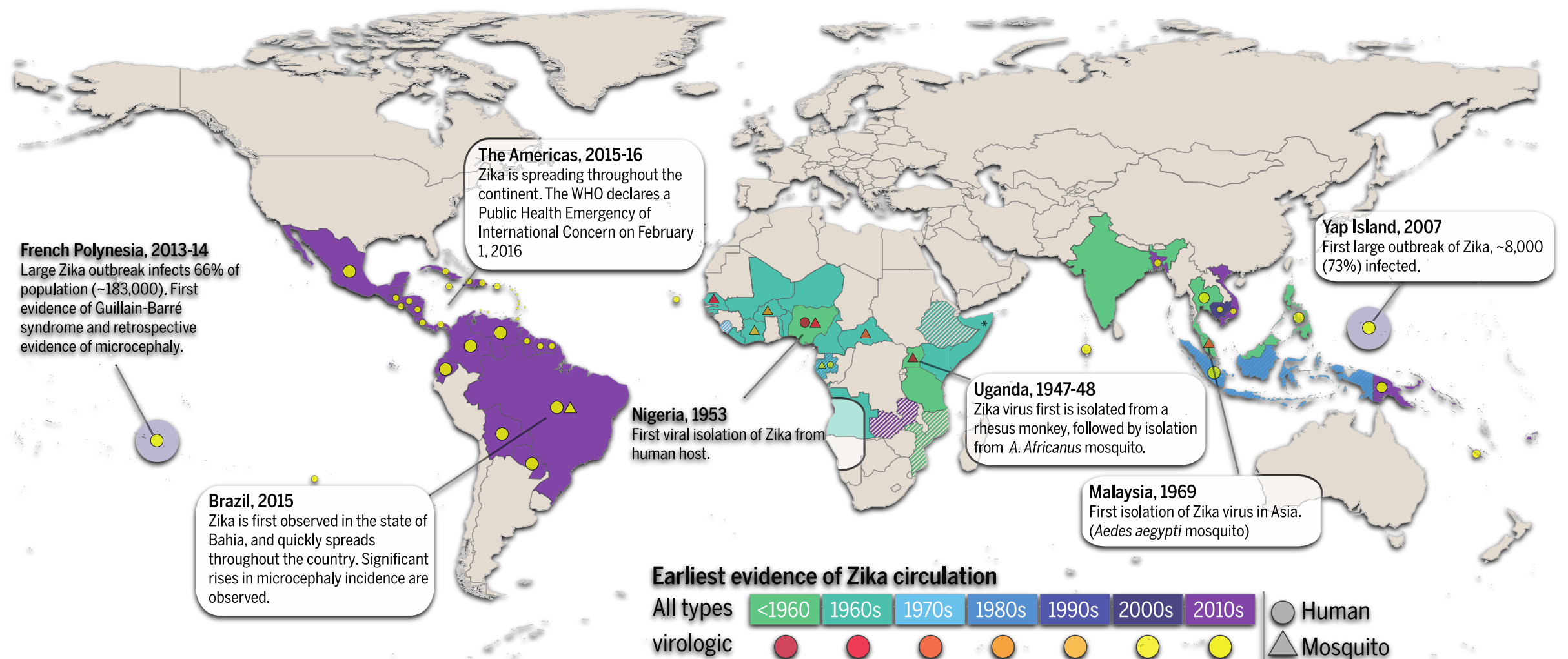
Vigilância Genômica do vírus CHIKV

Parte 3

Vigilância Genômica do vírus YFV

BREVE HISTÓRIA DE UMA EPIDEMIA

Perspectivas históricas são tipicamente obtidas através de estudos de serologia e não de estudos genéticos



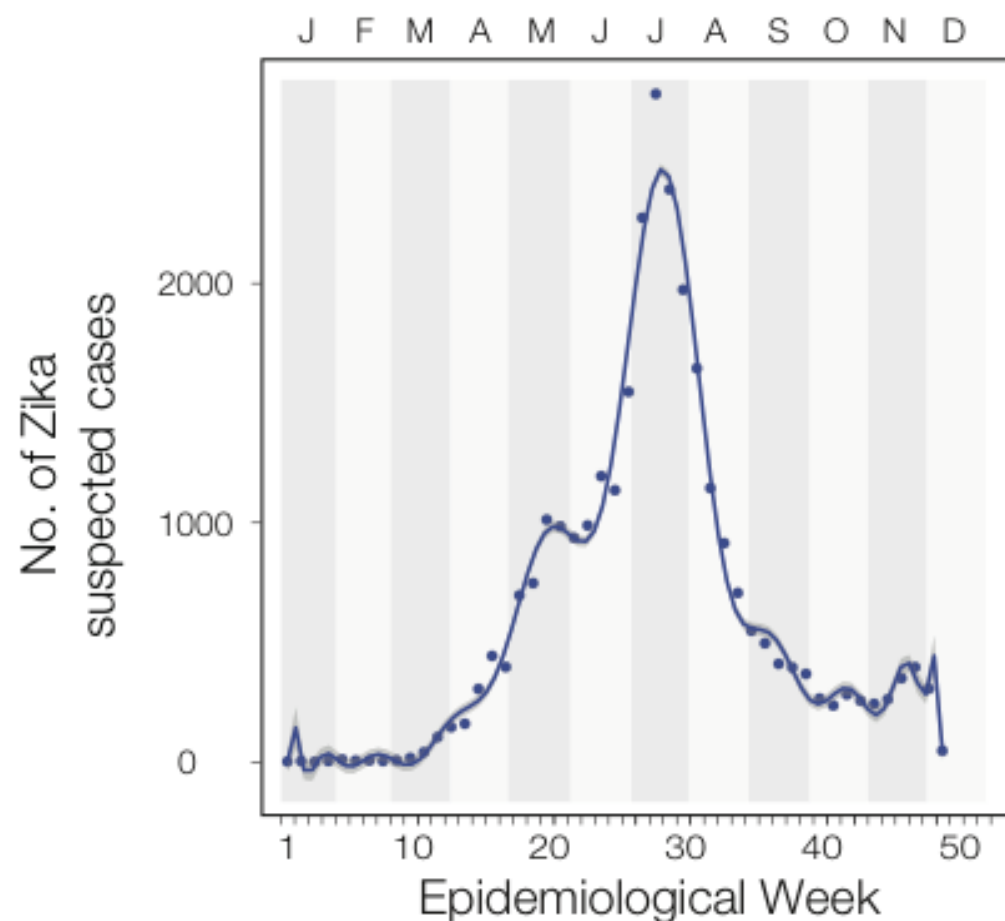
Associação entre Zika e microcefalia

Transmissão local confirmada em 29 Abril 2015

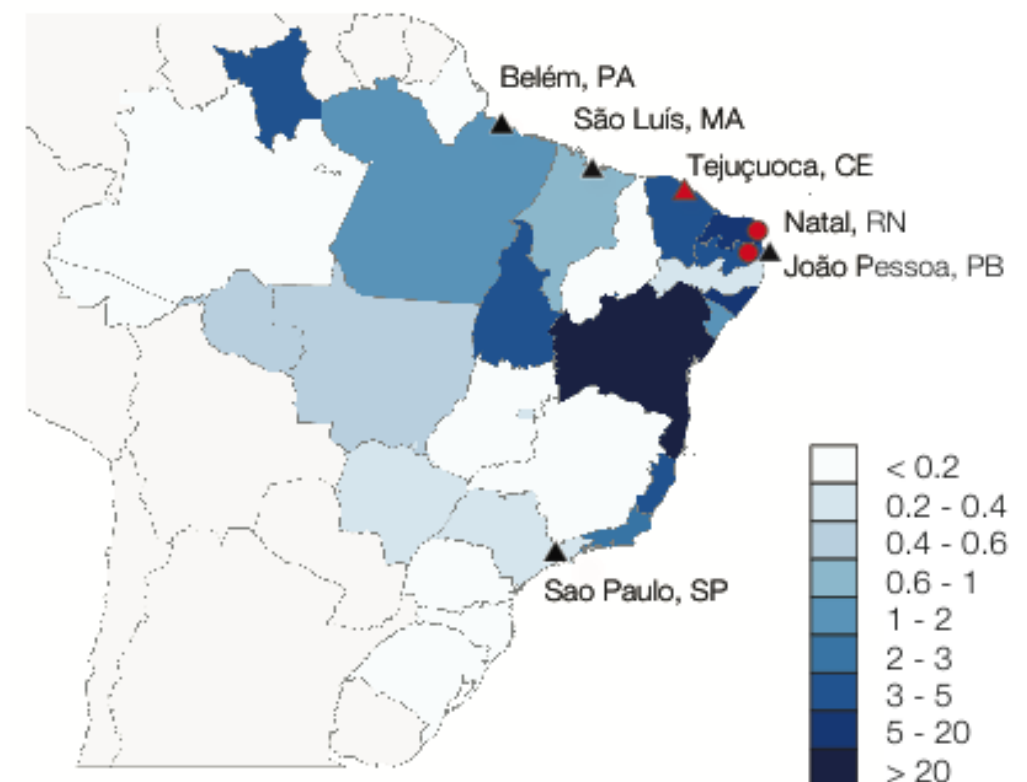
WHO Emergência Internacional de Saúde Pública: 01 Fev 2016

WHO/CDC confirma associação entre infecção com ZIKV e **microcefalia**: 13 Apr 2016

Pico epidémico em meados Julho 2015



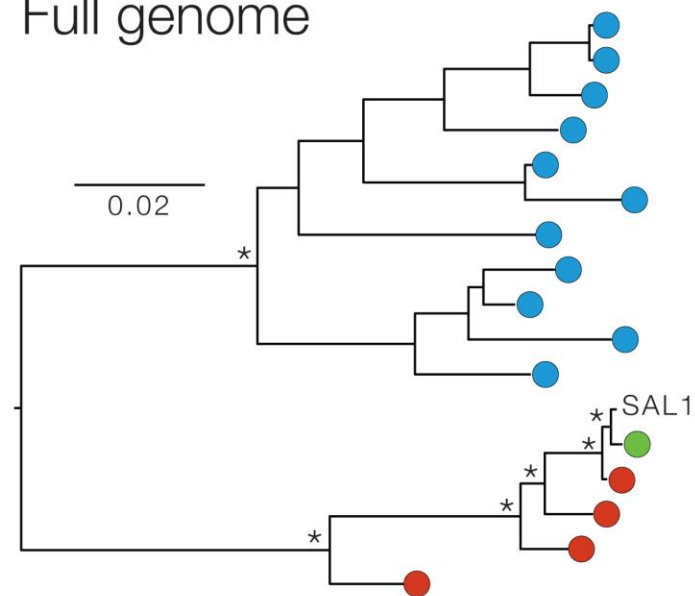
Incidência de ZIKV por 100,000 pessoas



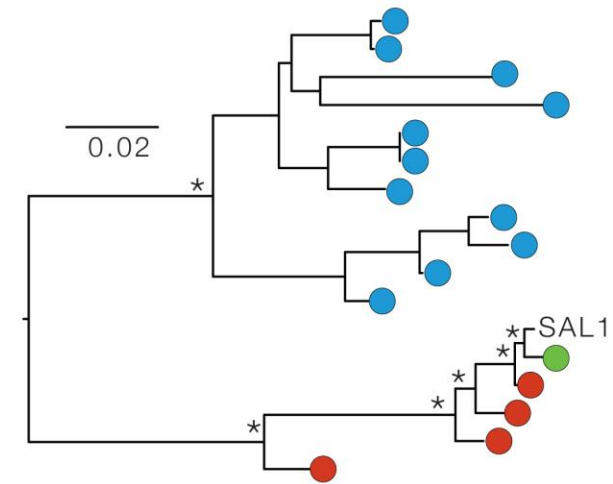


Genoma completo do vírus zika de Salvador, Bahia

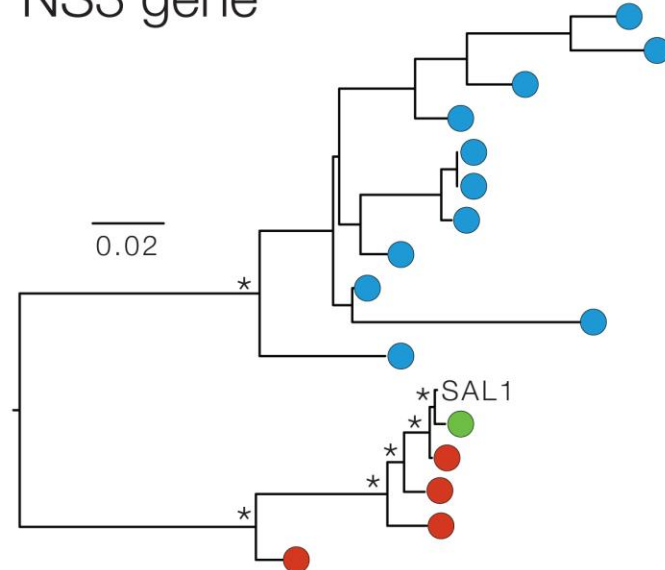
Full genome



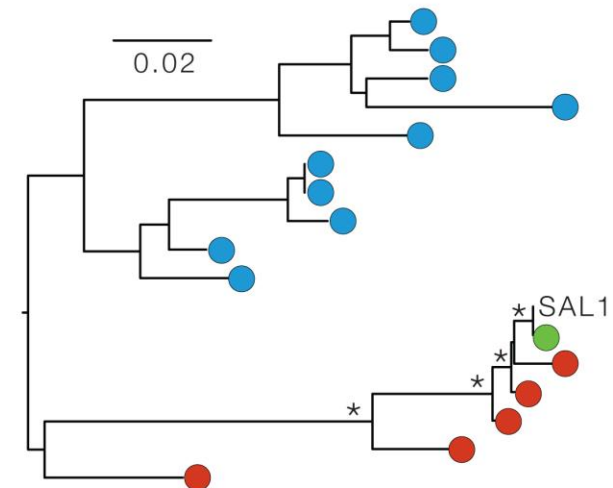
NS5 gene



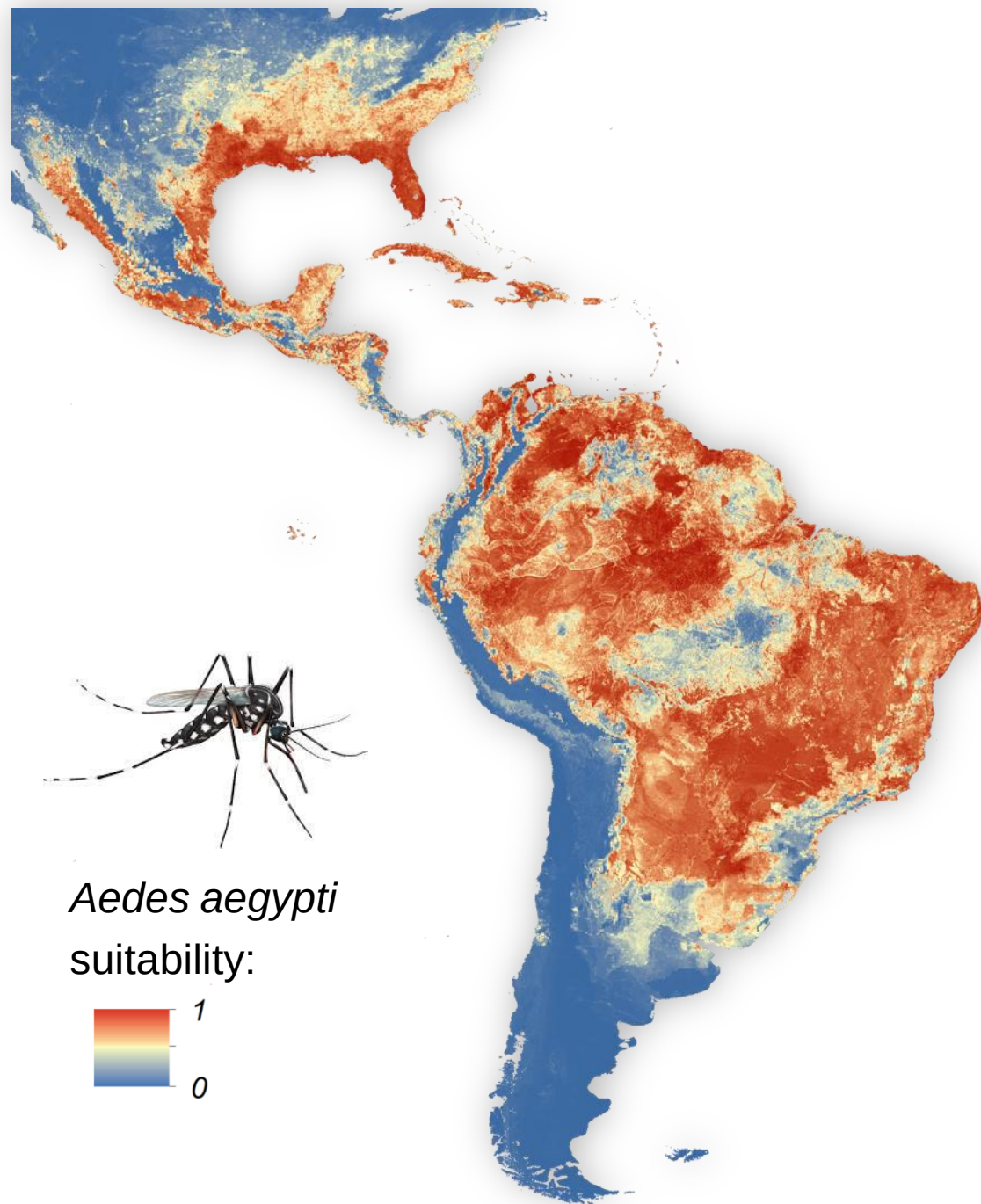
NS3 gene



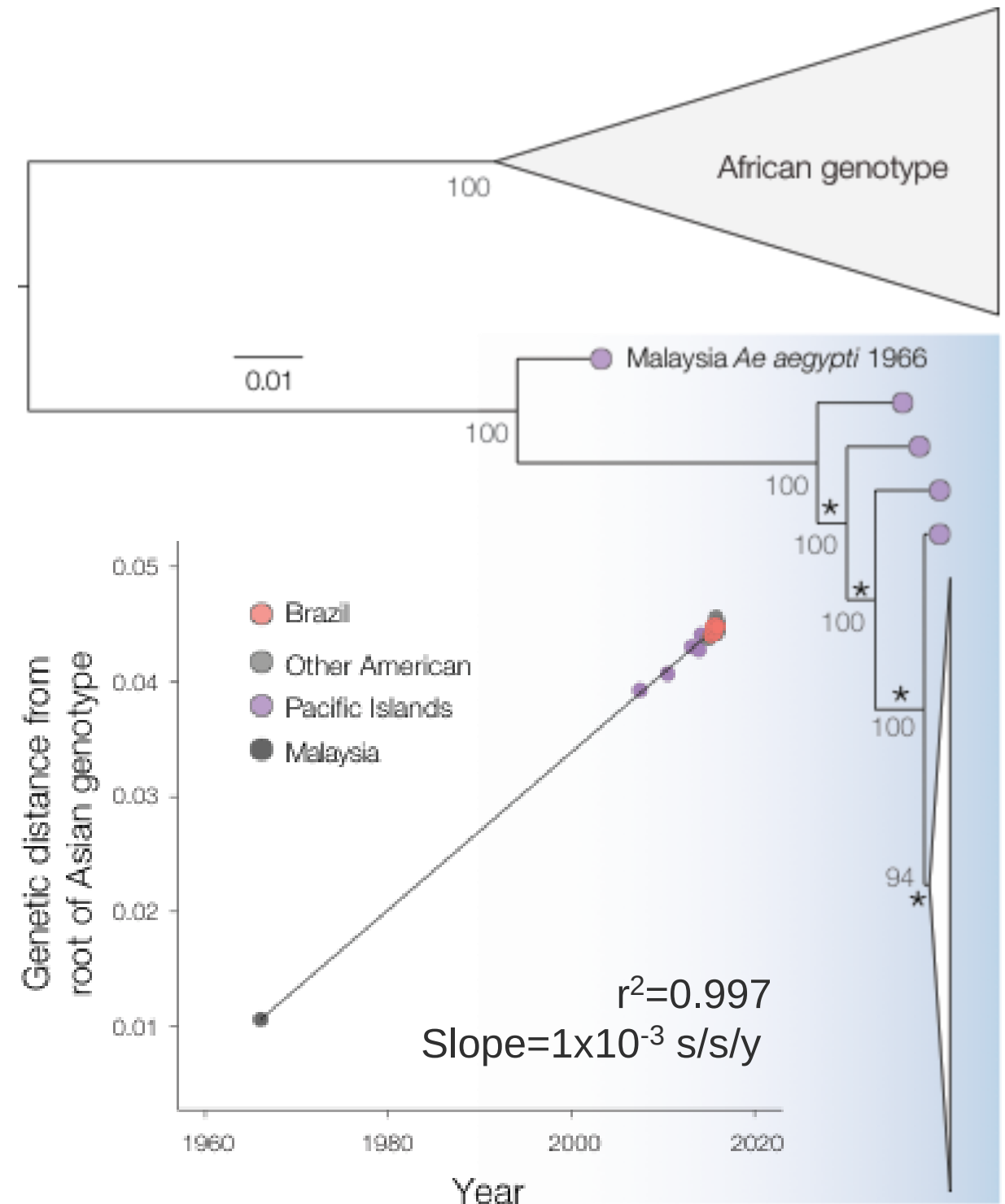
Env gene



Emergência do ZIKV foi resultado de adequabilidade climática, patogénese viral



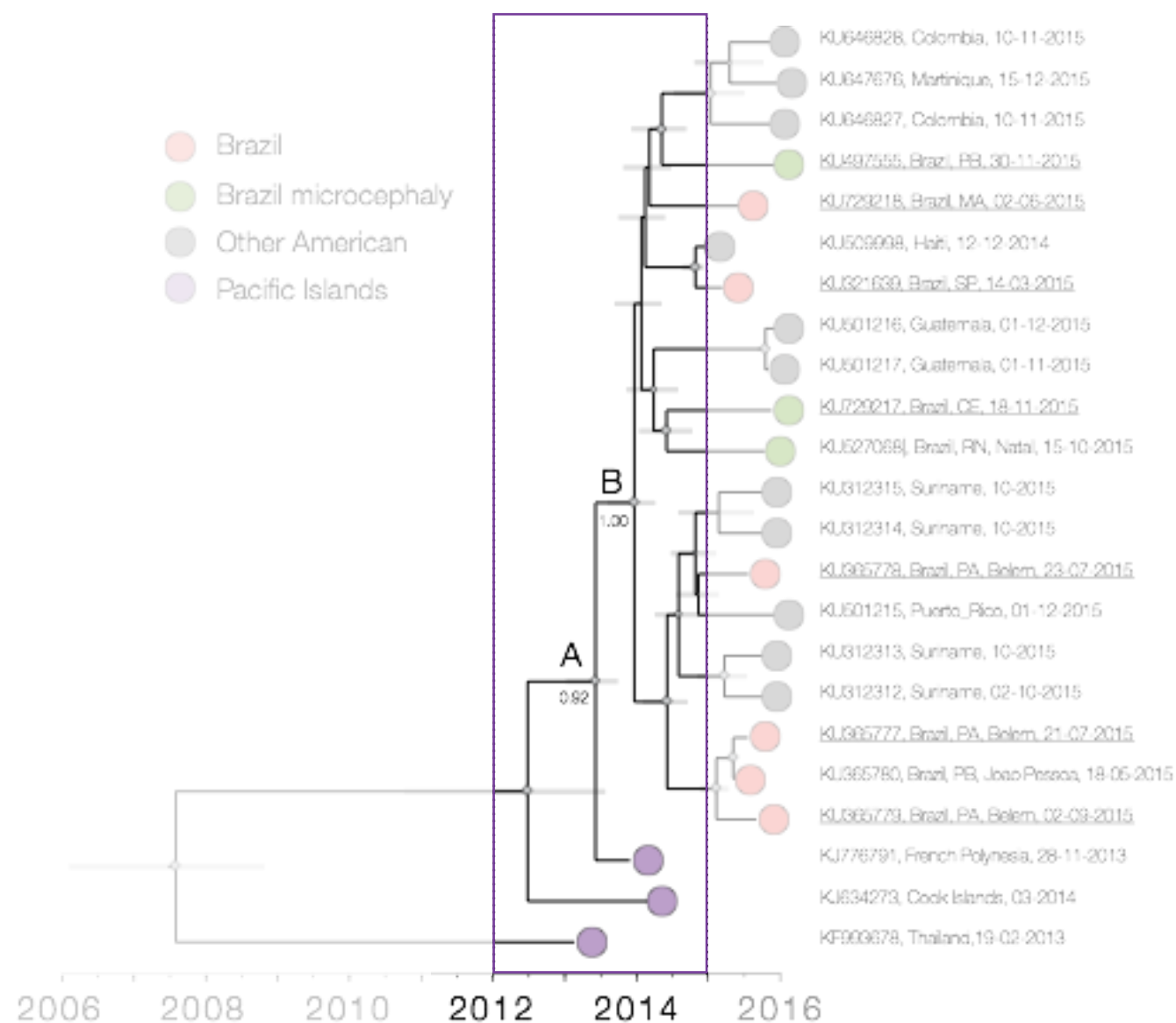
Kraemer MUG et al. *eLife* 2014



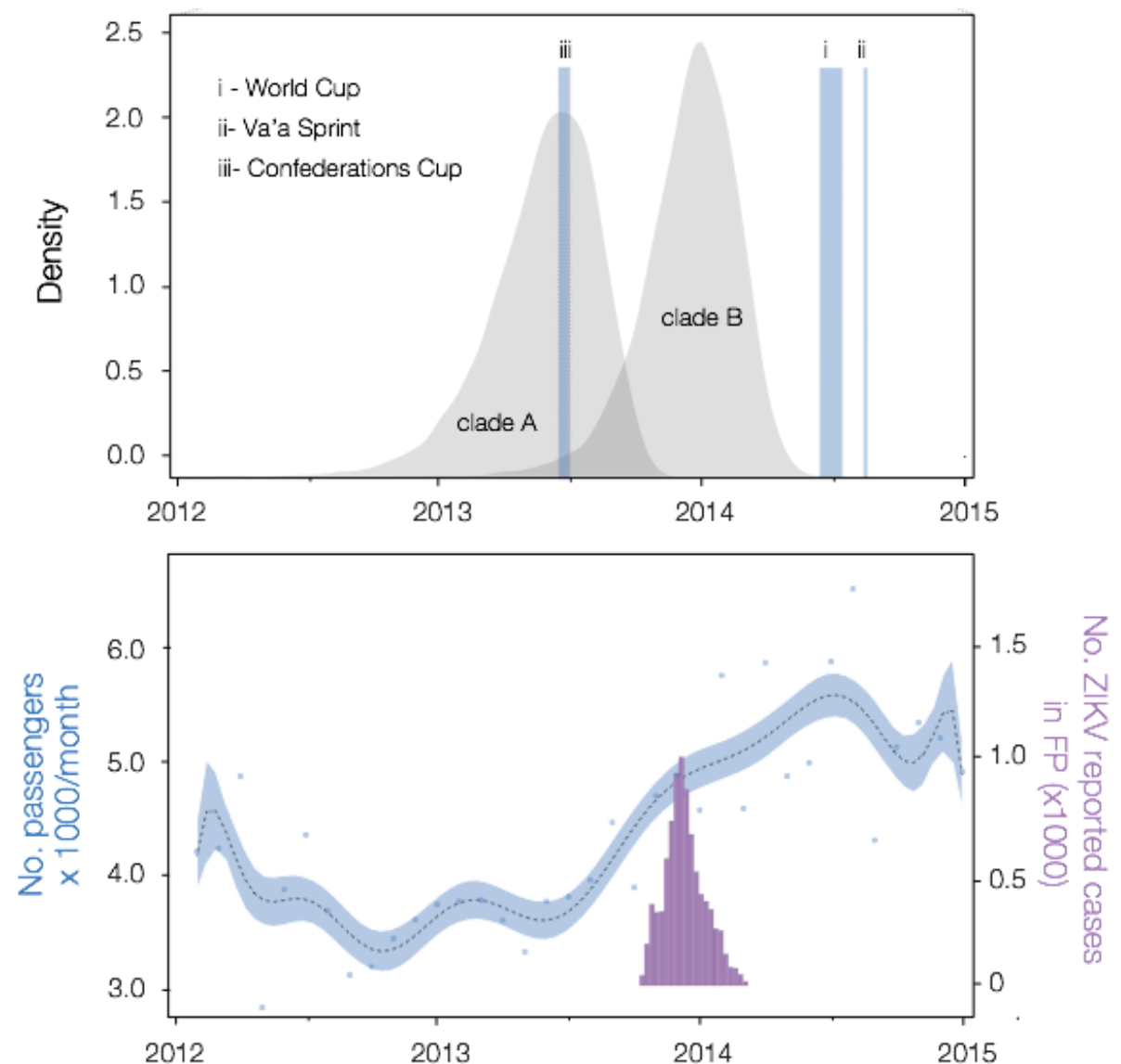
Faria et al. *Science* March 2016

Emergência do ZIKV foi resultado de adequabilidade climática, patogénese viral e **mobilidade global**

Genótipo ZIKV Asiático
filogenia relógio molecular



Combinação de dados
genéticos e de tráfego aéreo



Caracterização genética do ZIKV em diferentes fluidos corpóreos de um RN com agravos clínicos

A



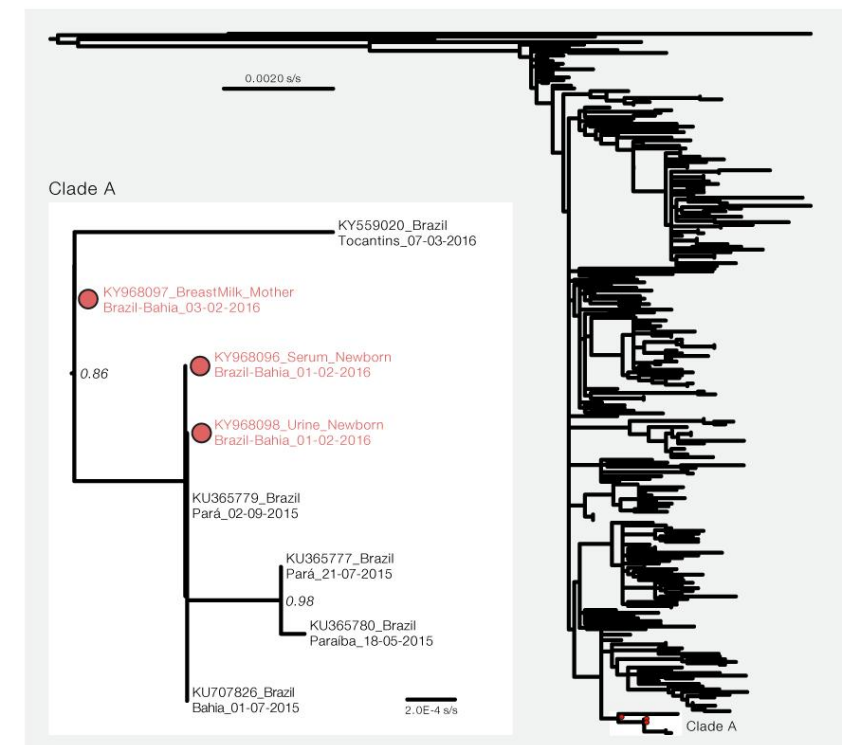
C.



B

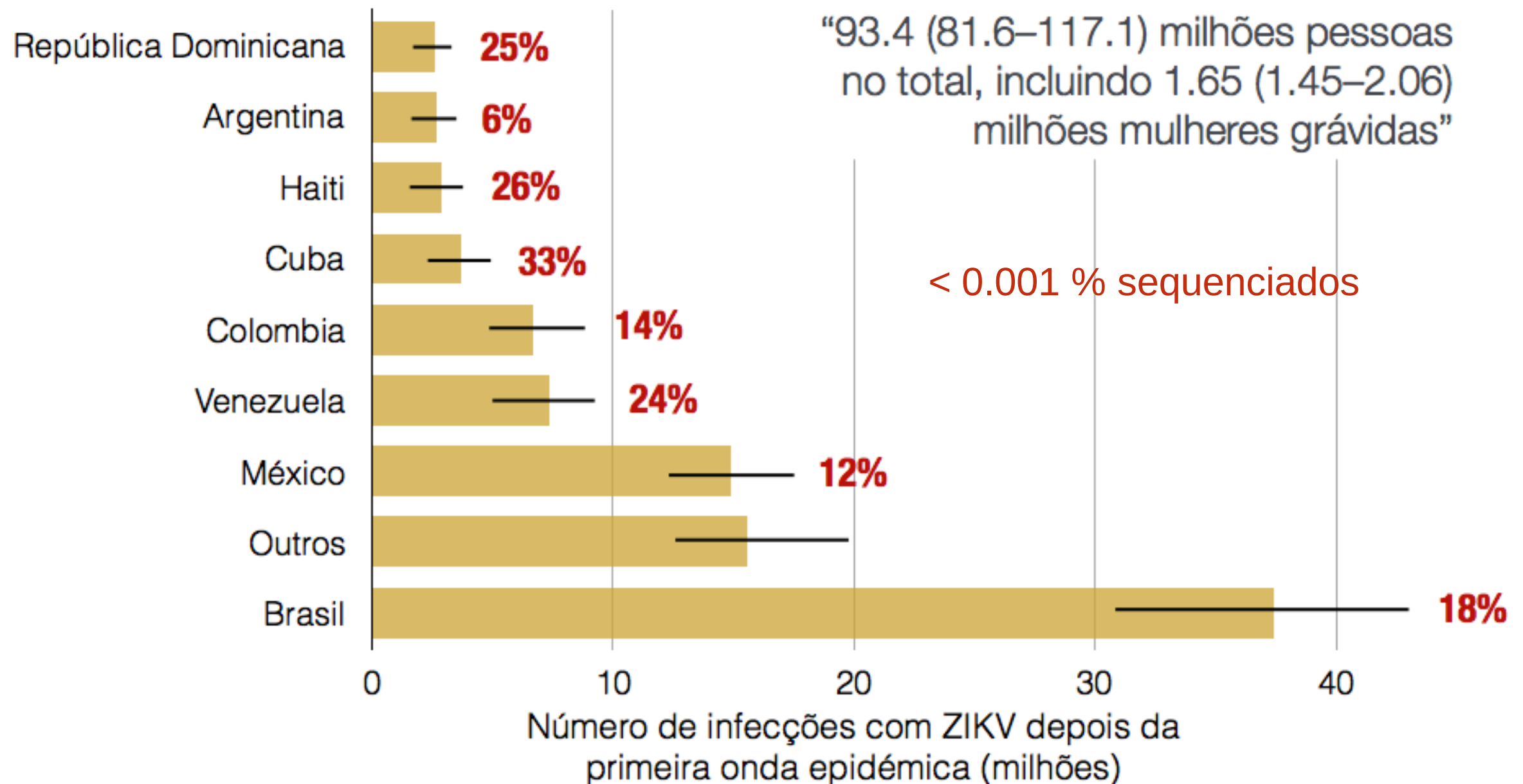


D.



Número real de infecções com ZIKV no Brasil cerca de 37 milhões

1 genoma : 1,5 milhões de casos



ZIKA

ZIKA IN BRAZIL REAL TIME ANALYSIS

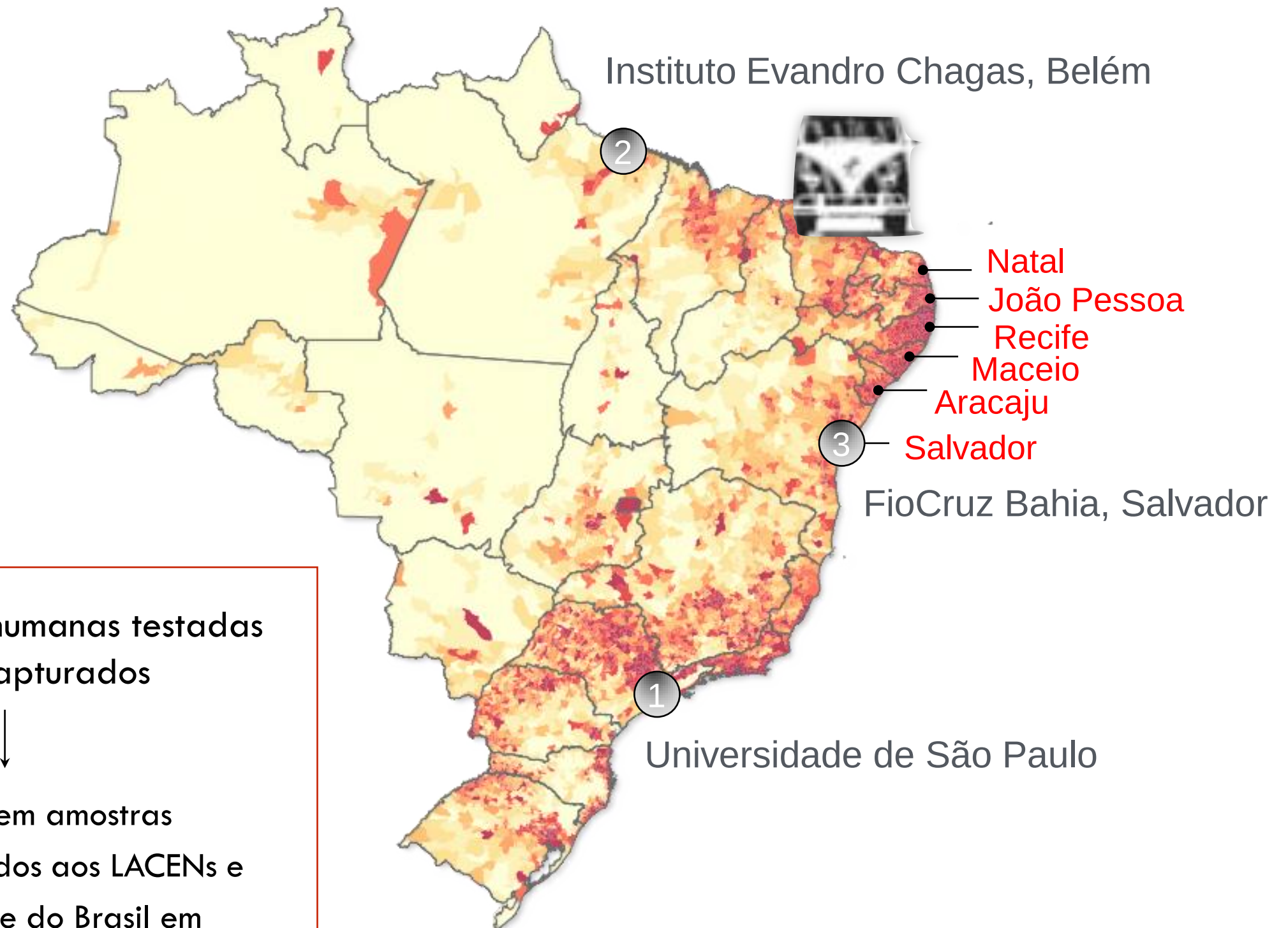


Nicholas Loman, University of Birmingham
Oliver Pybus e Nuno Faria, University of Oxford
Luiz Carlos Junior Alcantara, FIOCRUZ-RJ
Márcio Nunes, IEC-Pará
Ester Sabino, USP-SP



Projecto ZiBRA

Zika in Brazil real-time analysis



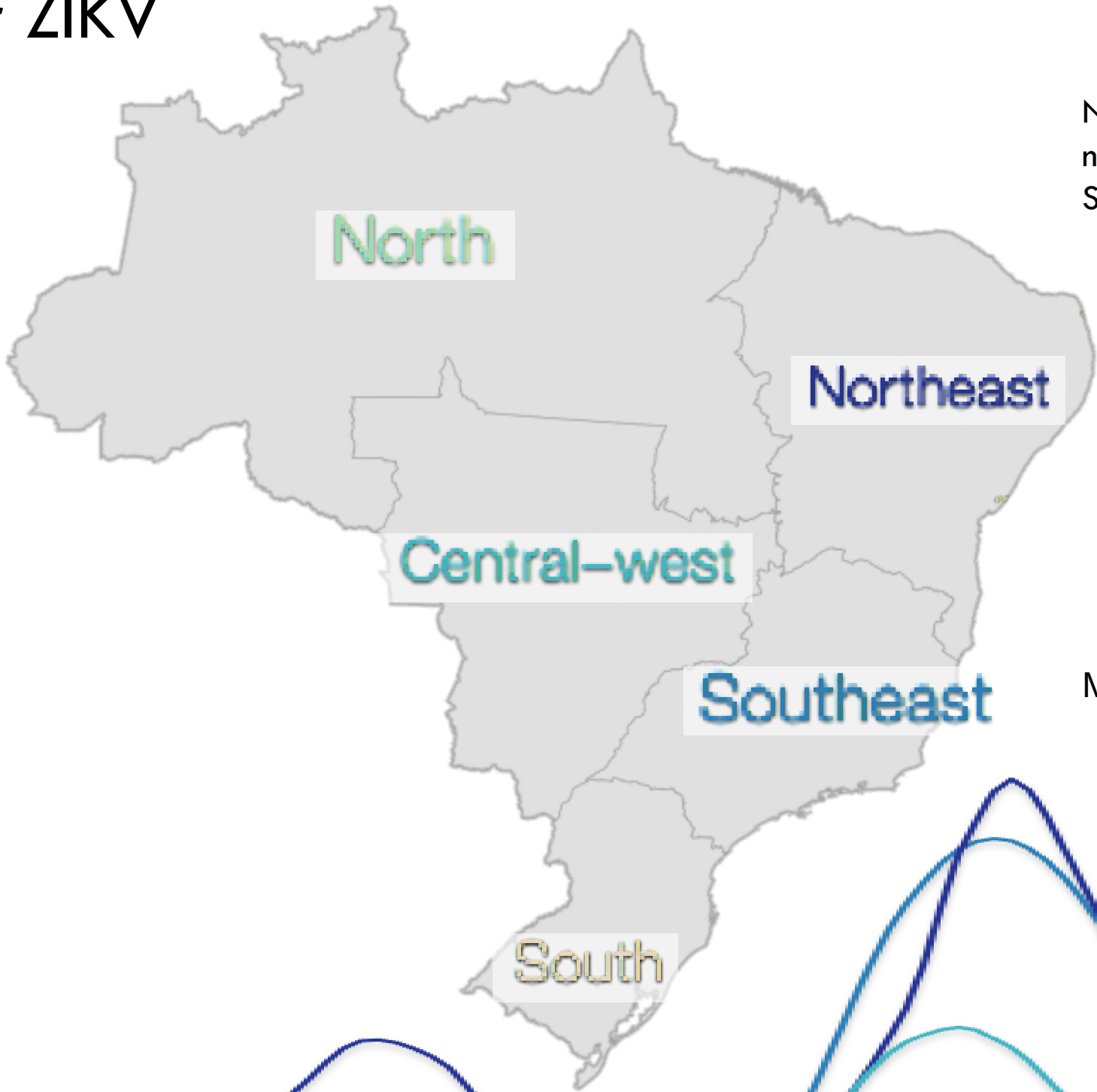
1401 amostras humanas testadas
838 mosquitos capturados



RT-PCR resultados em amostras
humanas comunicados aos LACENs e
Ministério da Saúde do Brasil em

48h

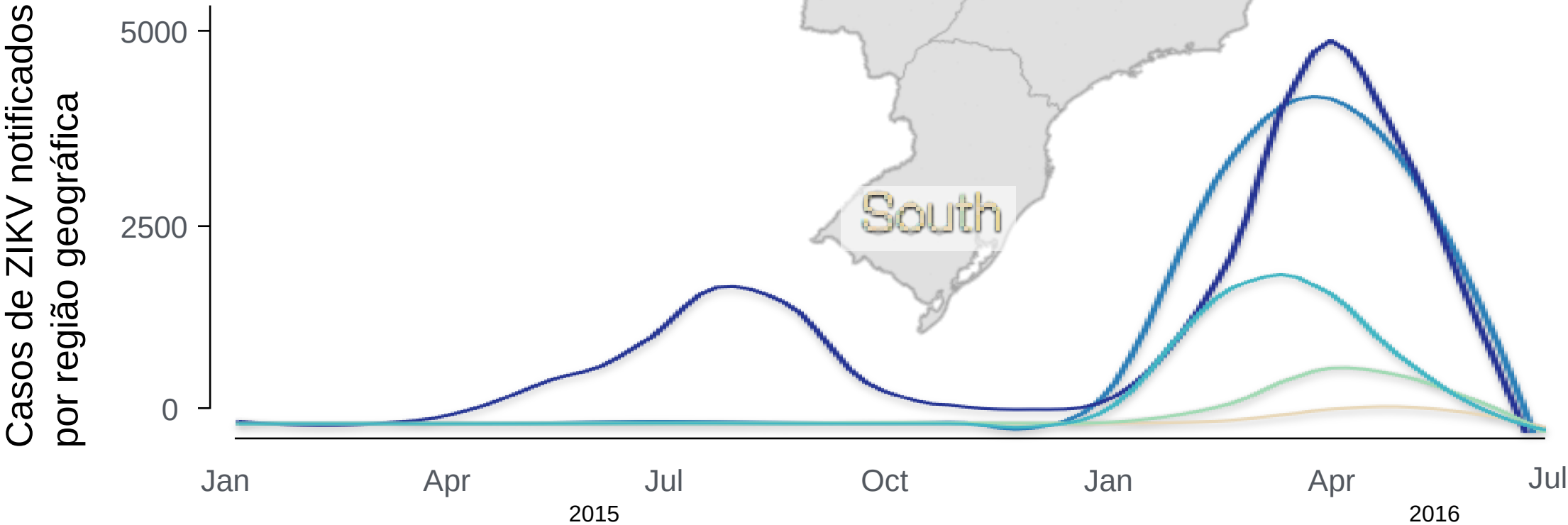
Nordeste foi a região mais afectada por ZIKV



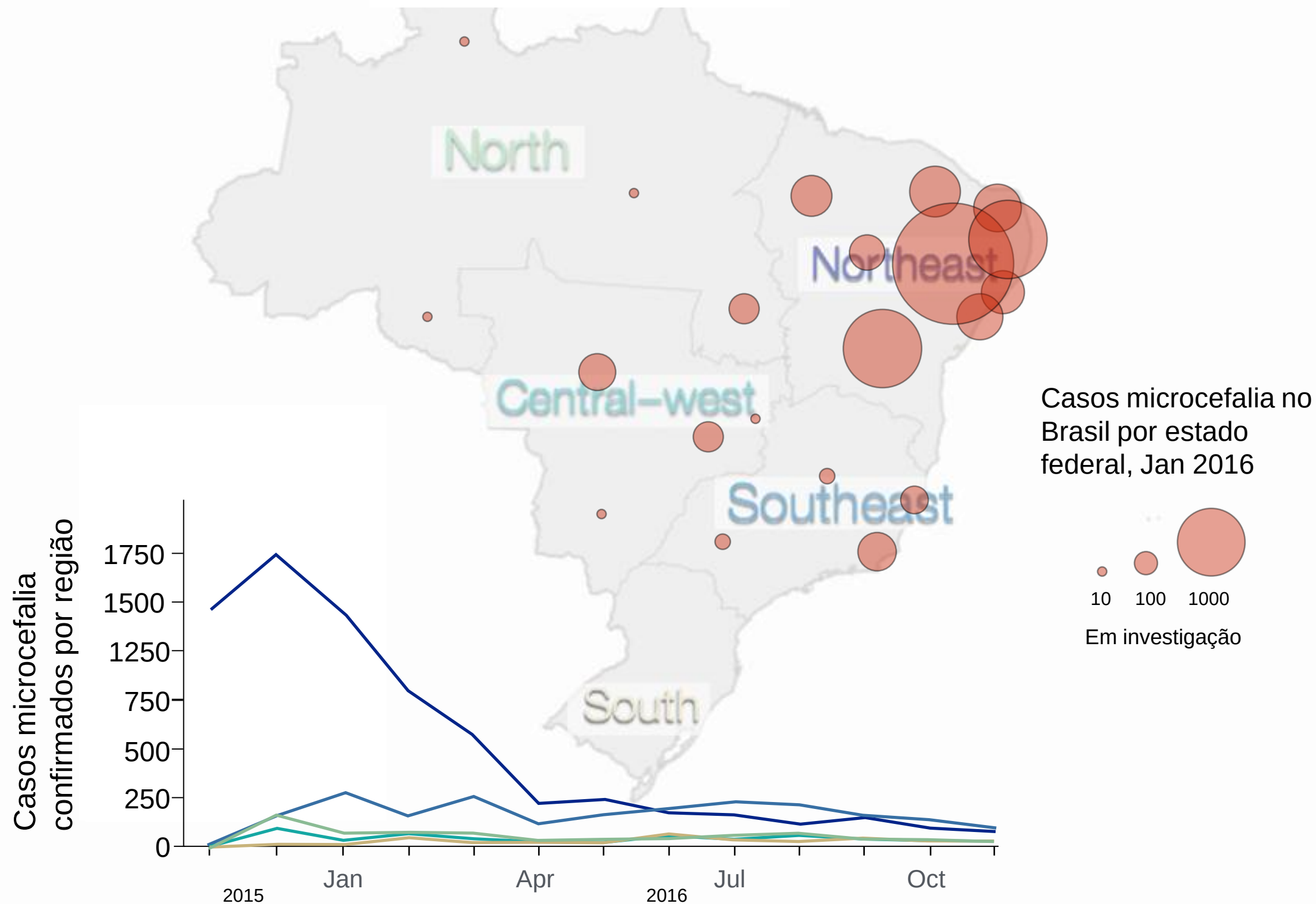
Número de casos de ZIKV notificados ao Ministério da Saúde por município:

- <1
- 1-30
- 30-100
- 100-1000
- >1000

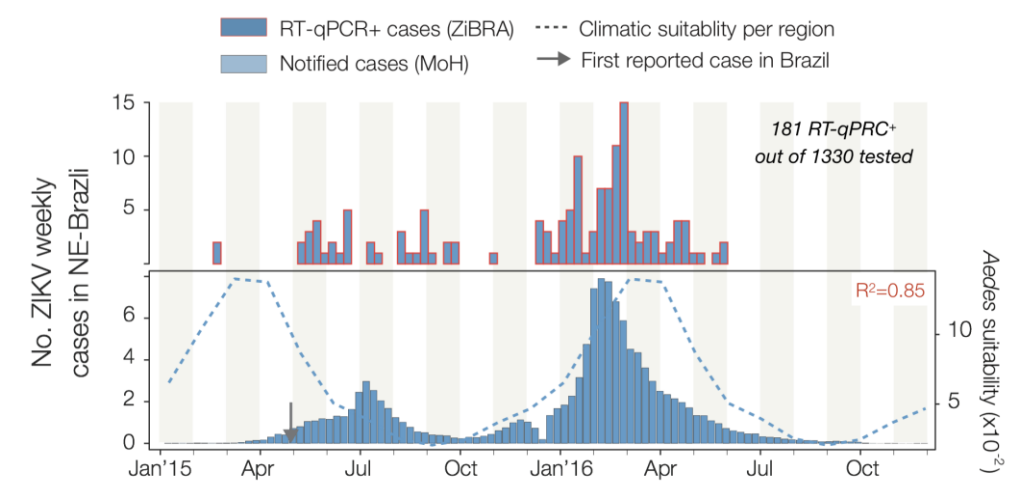
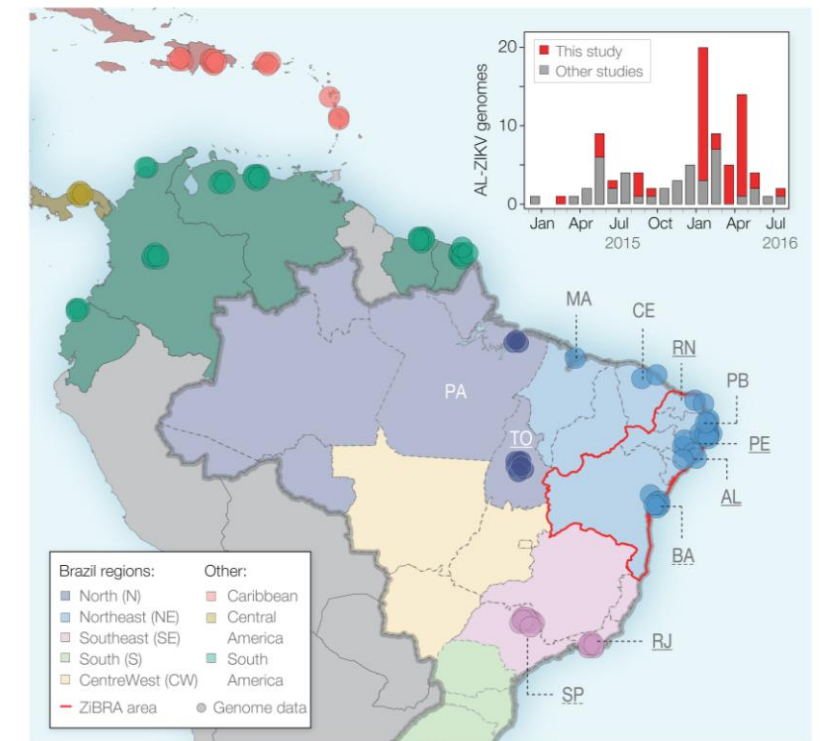
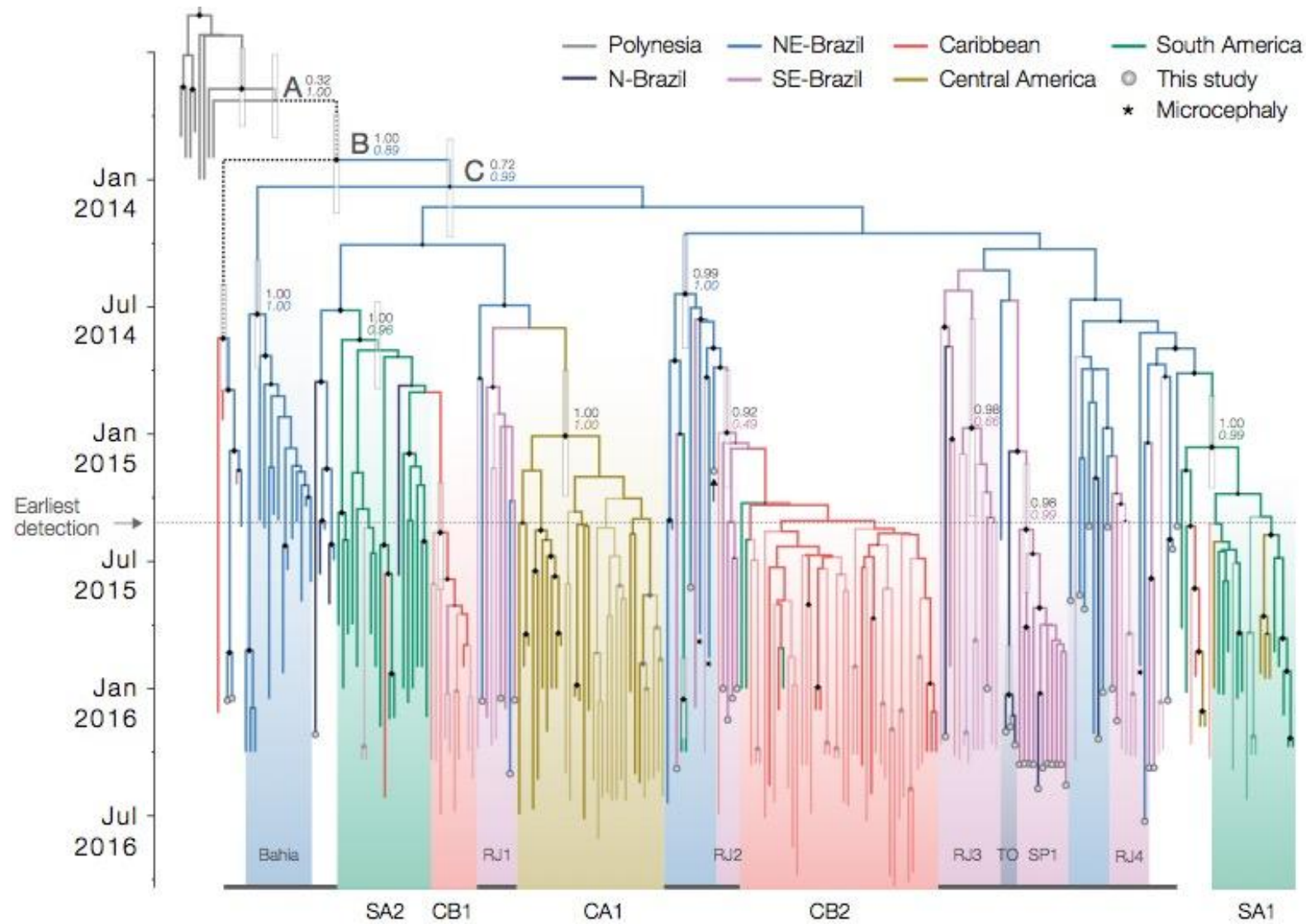
Mapa por Jose Lourenço



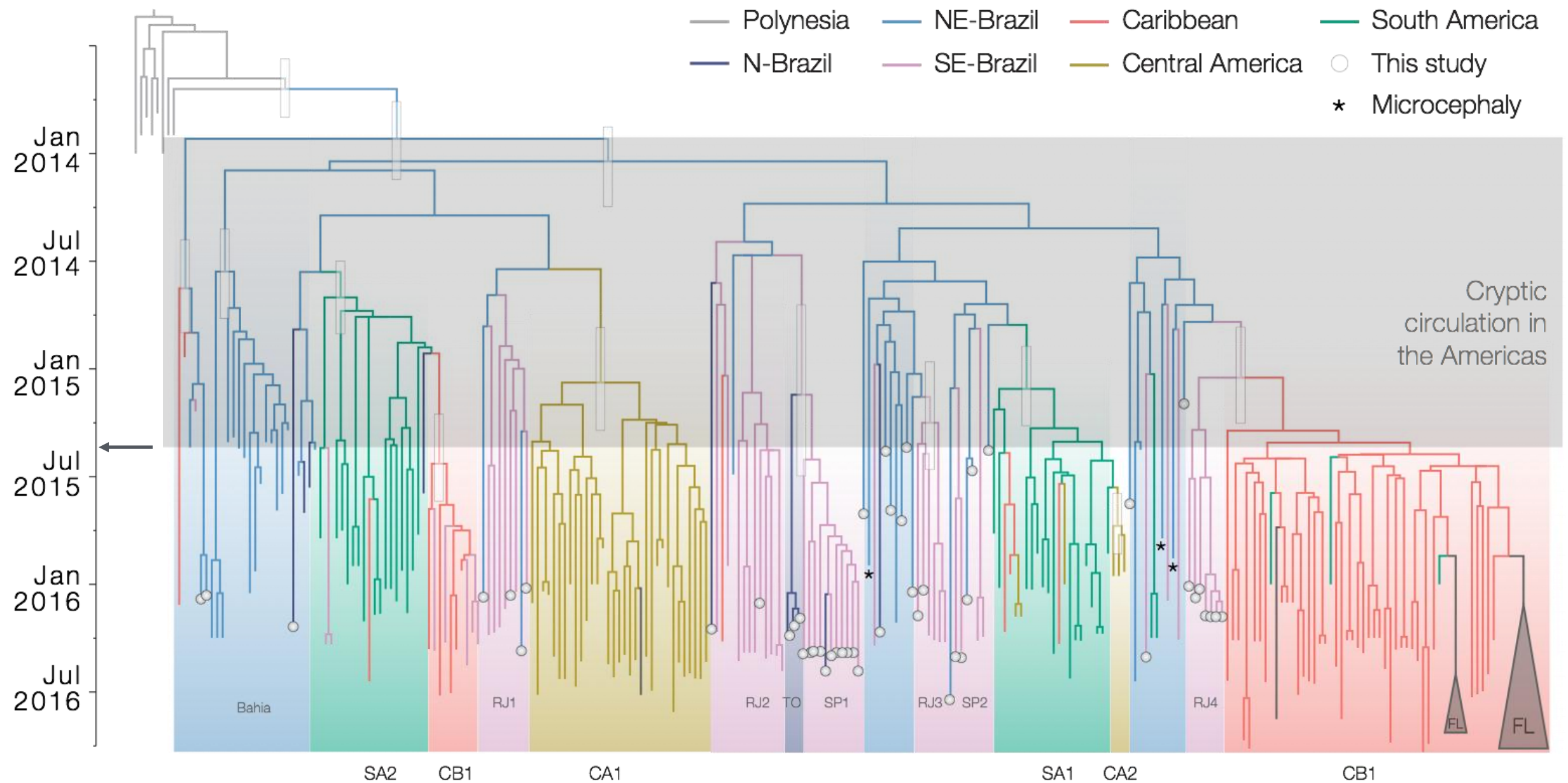
Nordeste foi a região mais
afectada por ZIKV e por microcefalia



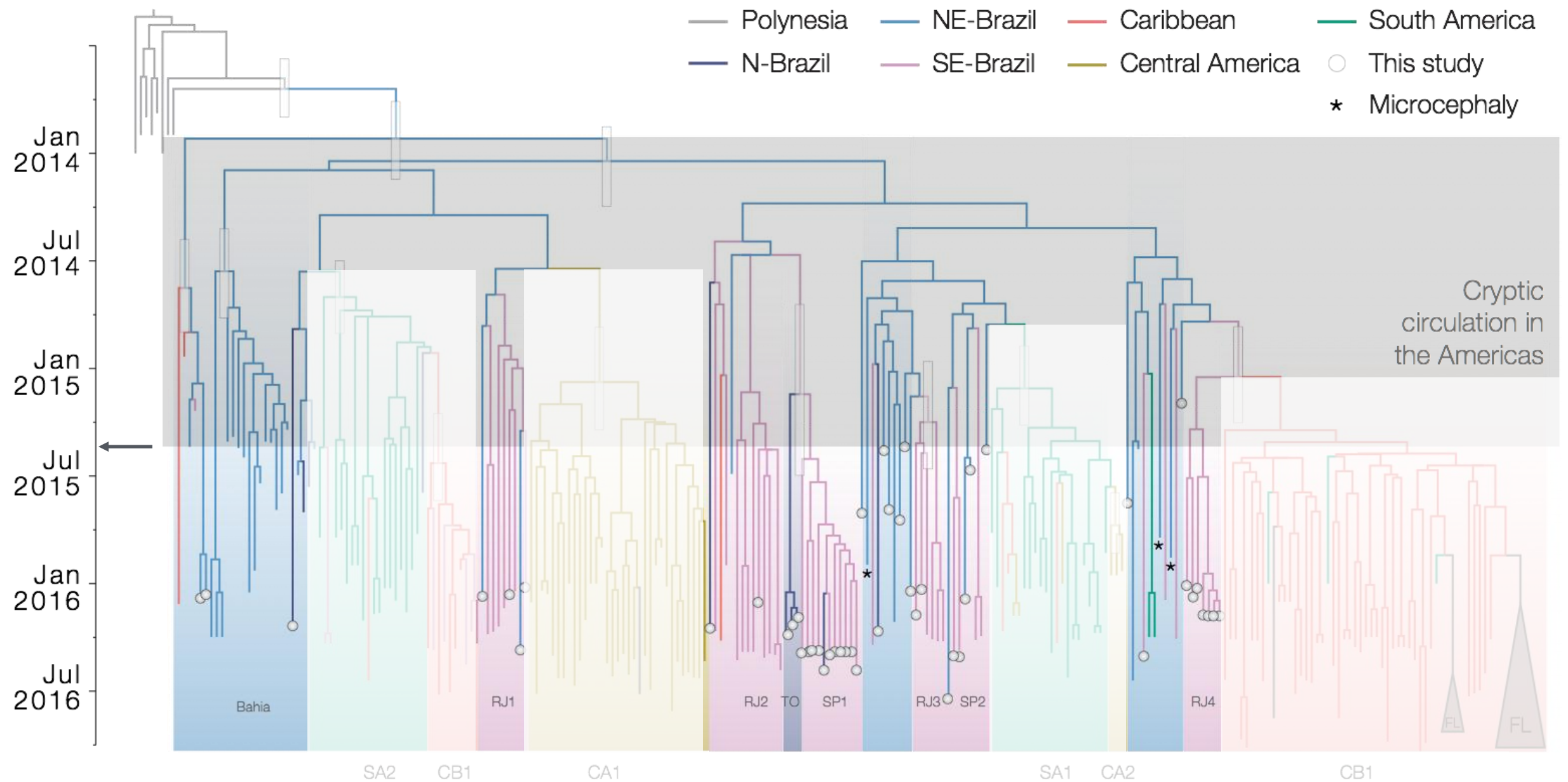
54 Genomas do vírus zika obtidos durante uma “road trip” no nordeste do Brasil



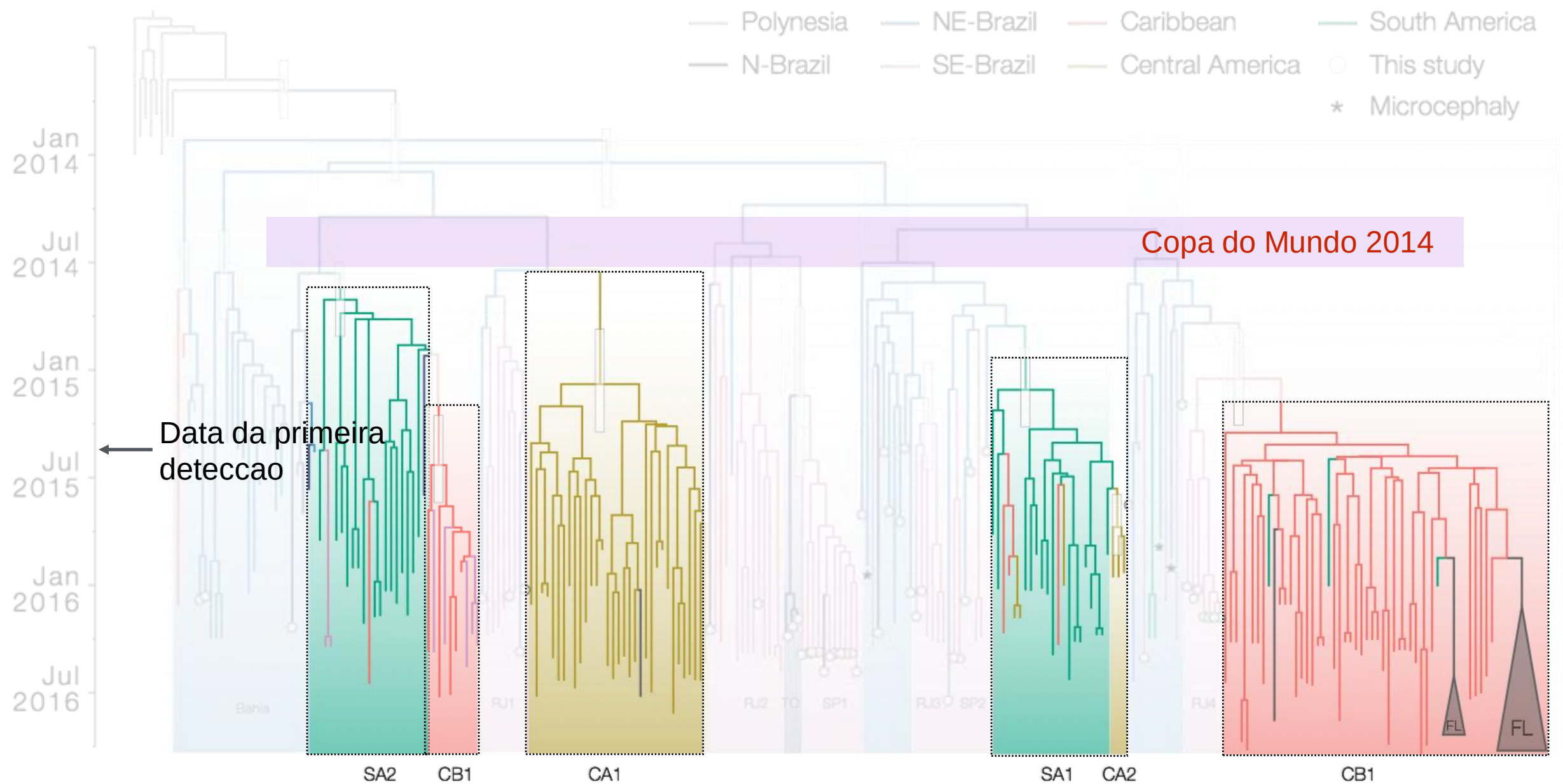
54 Genomas do vírus zika obtidos durante uma «road trip» no nordeste do Brasil



54 Genomas do vírus zika obtidos durante uma “road trip” no nordeste do Brasil

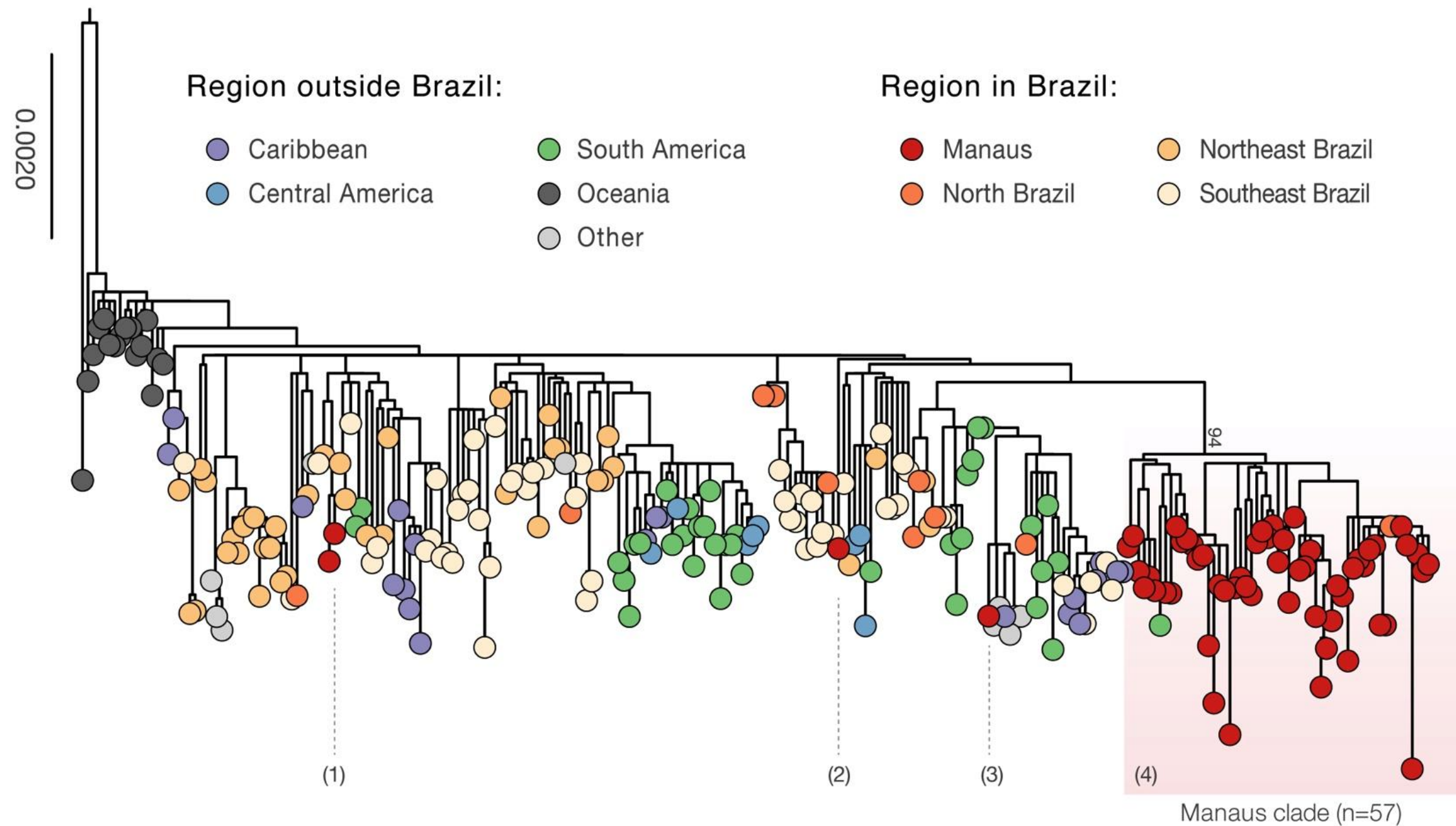


54 Genomas do vírus zika obtidos durante uma «road trip» no nordeste do Brasil



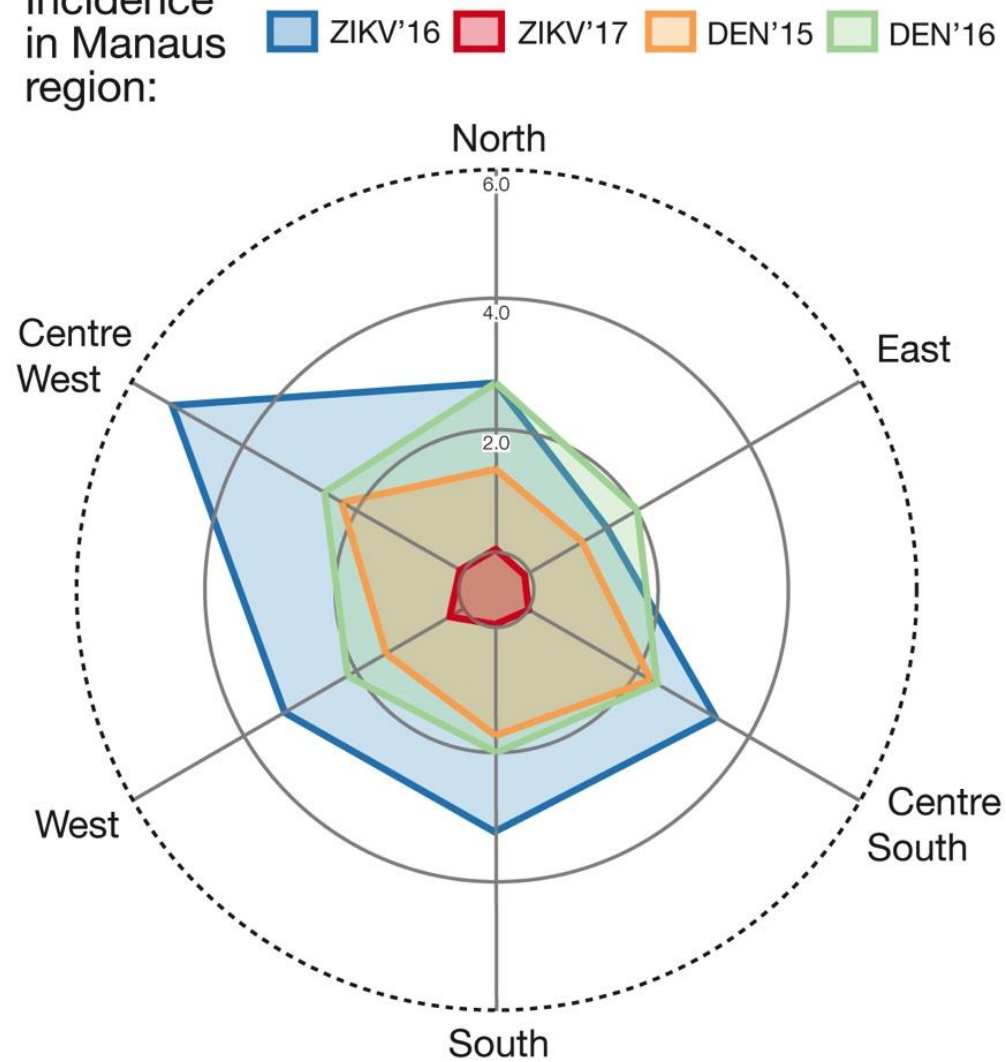
Vigilância genômica na região Amazônica

Persistência local do vírus Zika na região Norte (2015 – 2017)

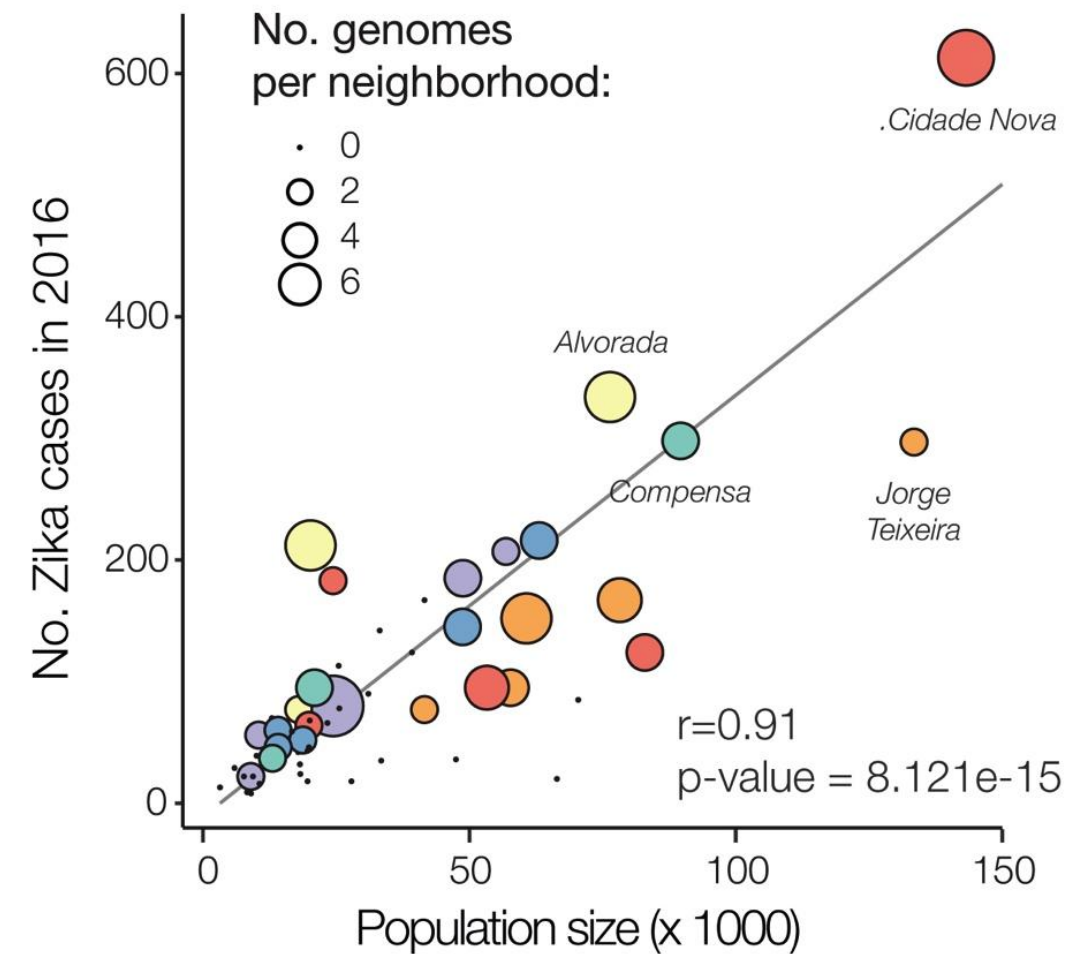


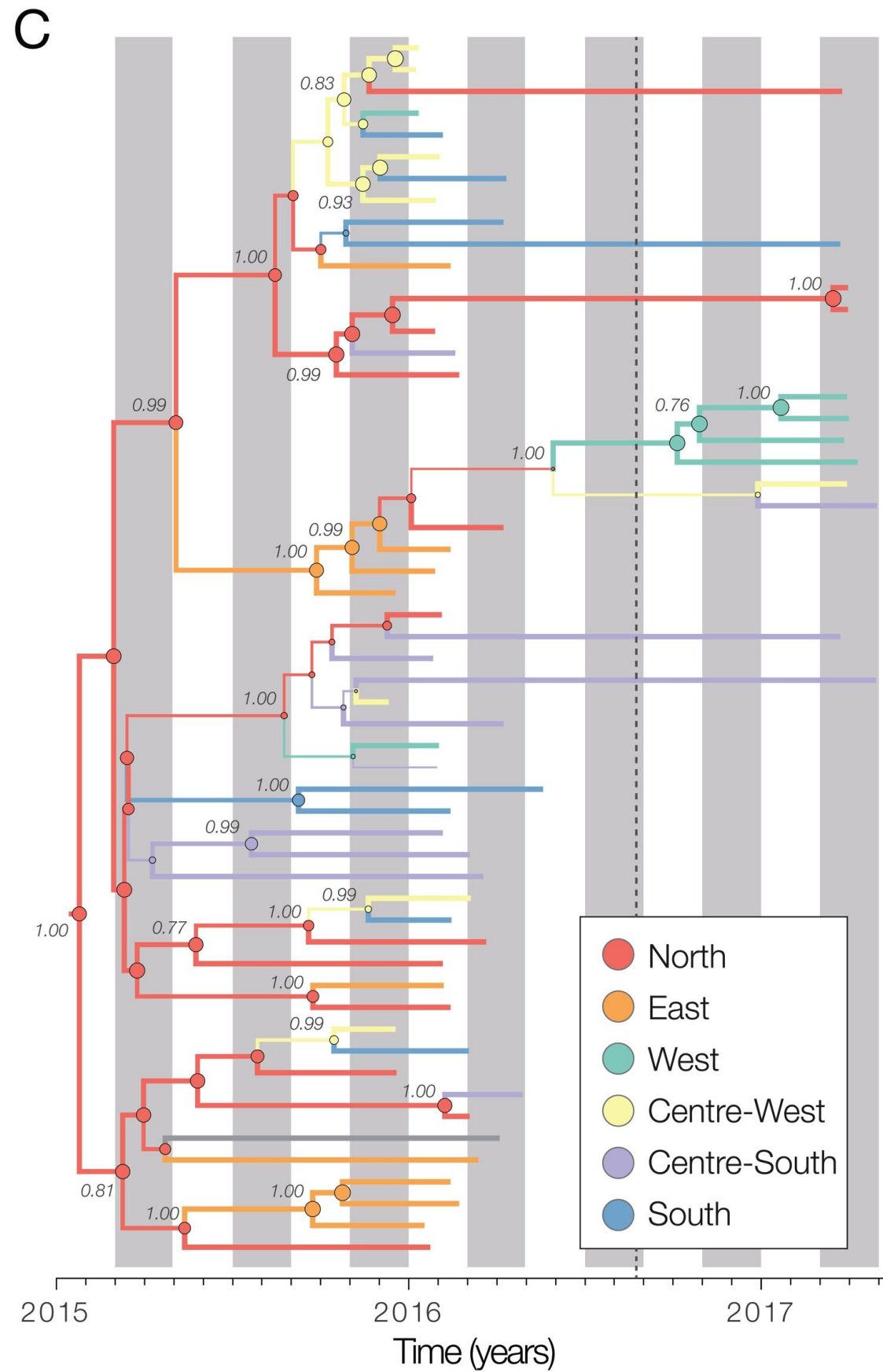
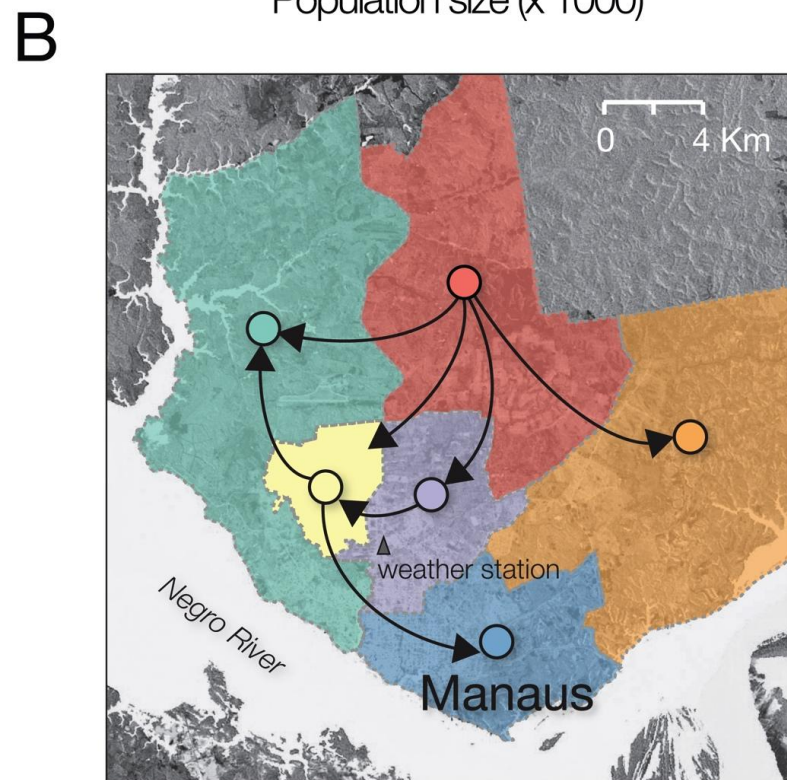
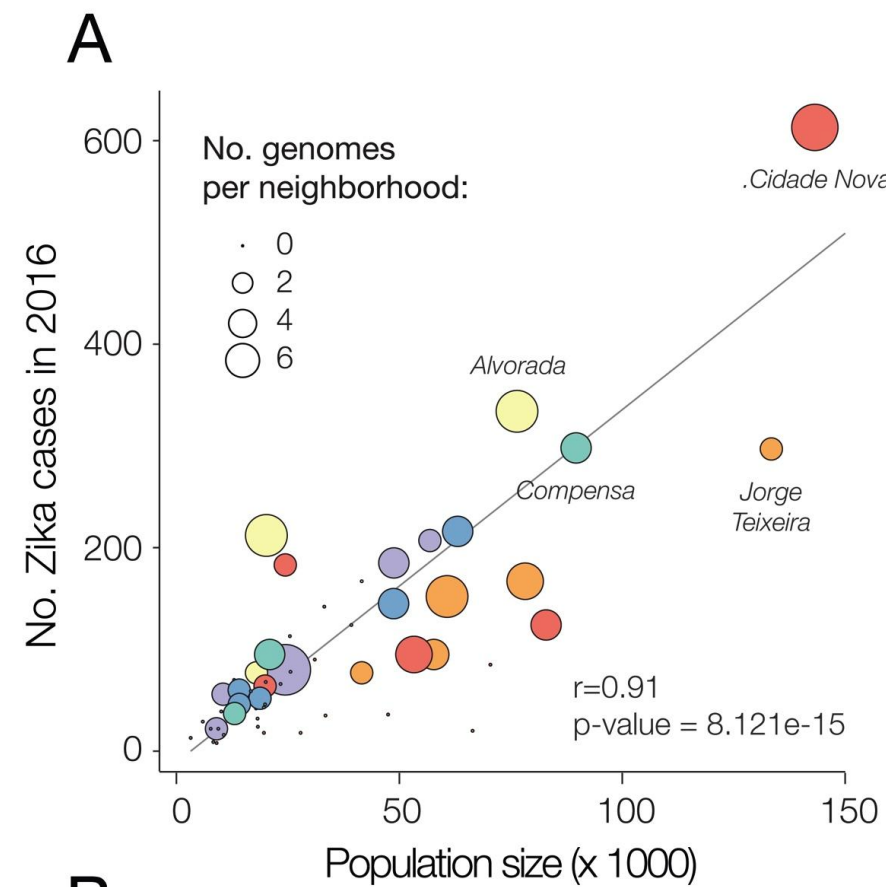
Combinação de dados genômicos + epidemiológicos + ambientais

A Incidence
in Manaus
region:



B





Parte 1

Vigilância Genômica do vírus ZIKV

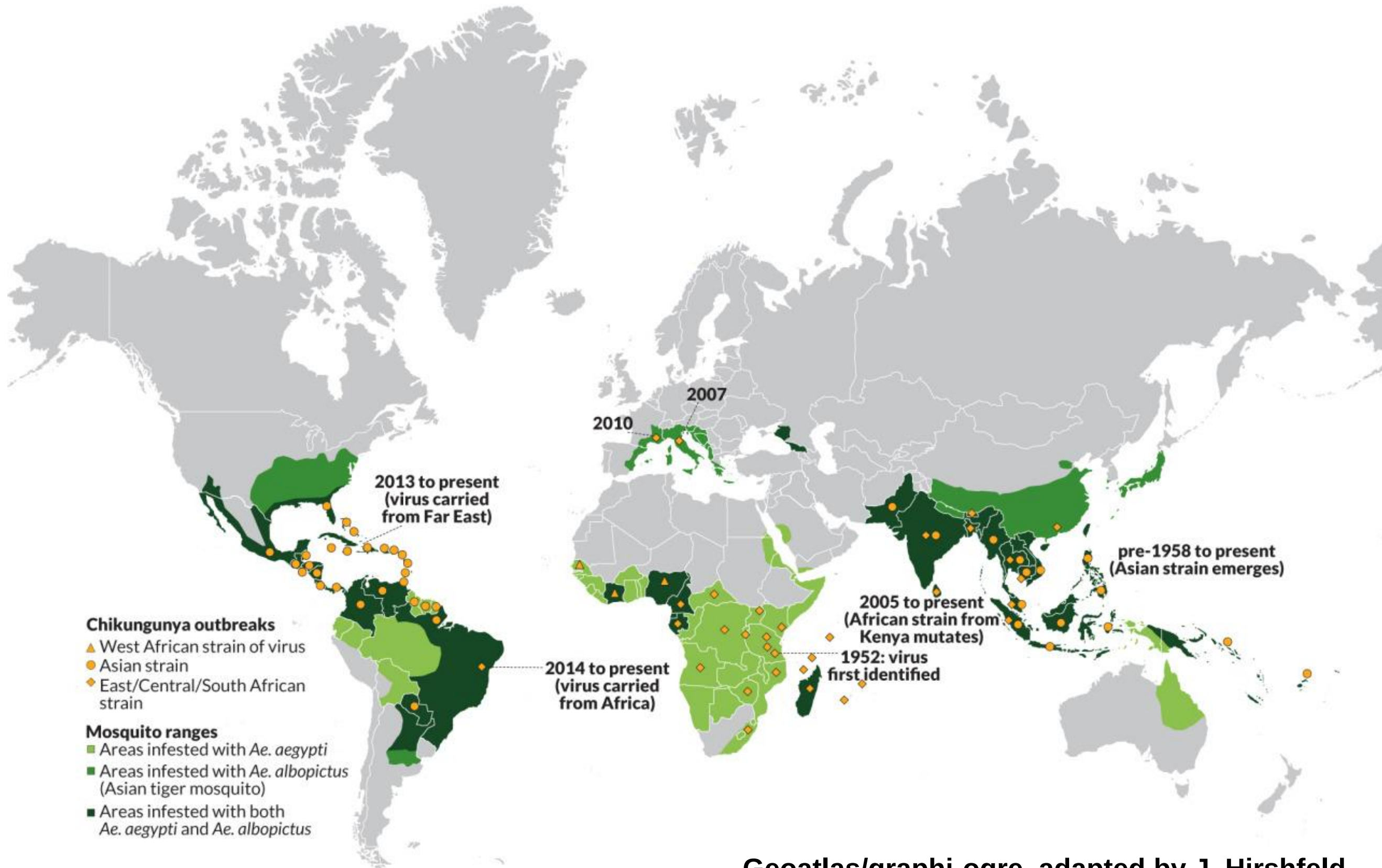
Parte 2

Vigilância Genômica do vírus CHIKV

Parte 3

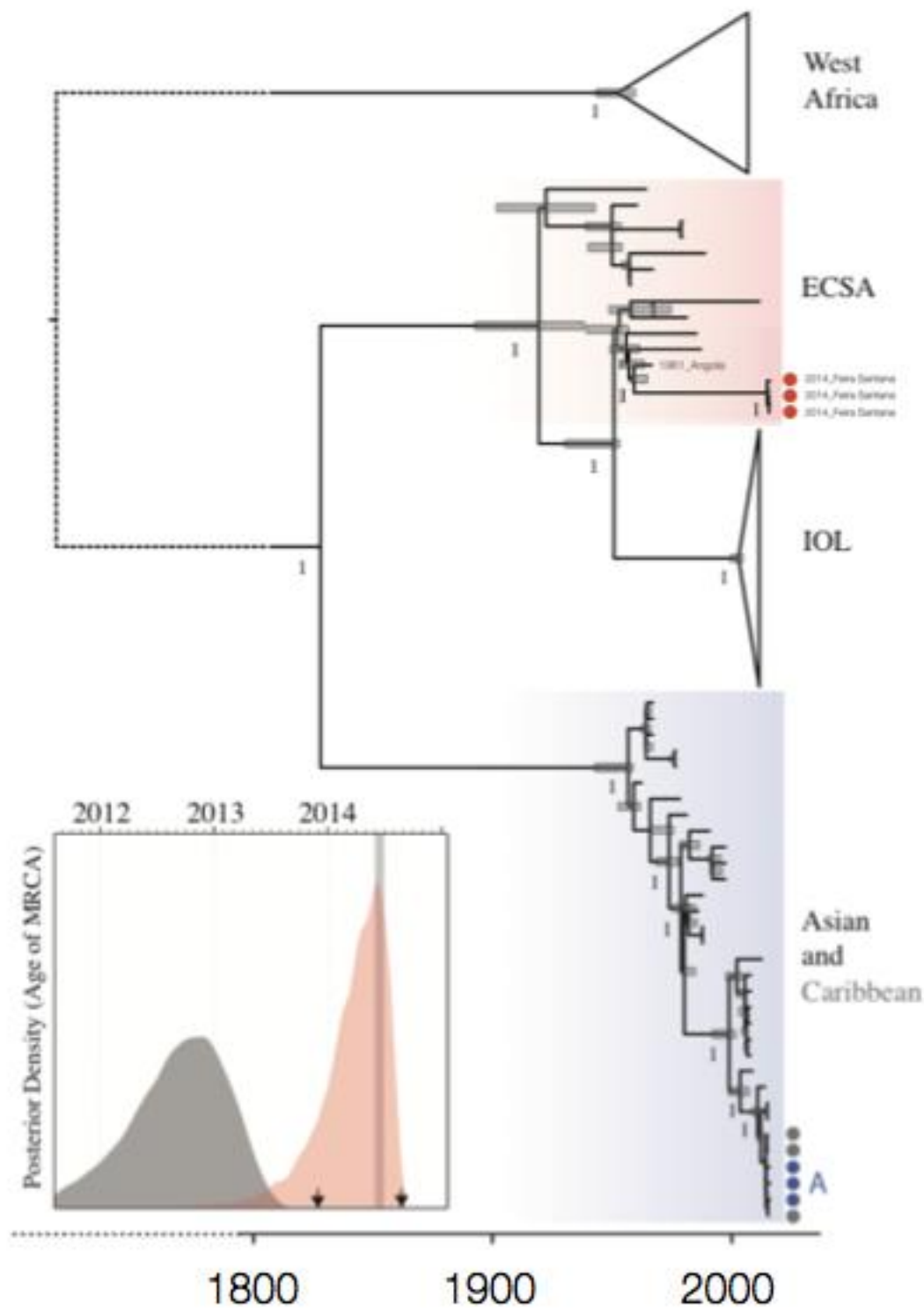
Vigilância Genômica do vírus YFV

BREVE HISTÓRIA DE UMA EPIDEMIA





Co-circulação de múltiplos arbovirus no território Brasileiro

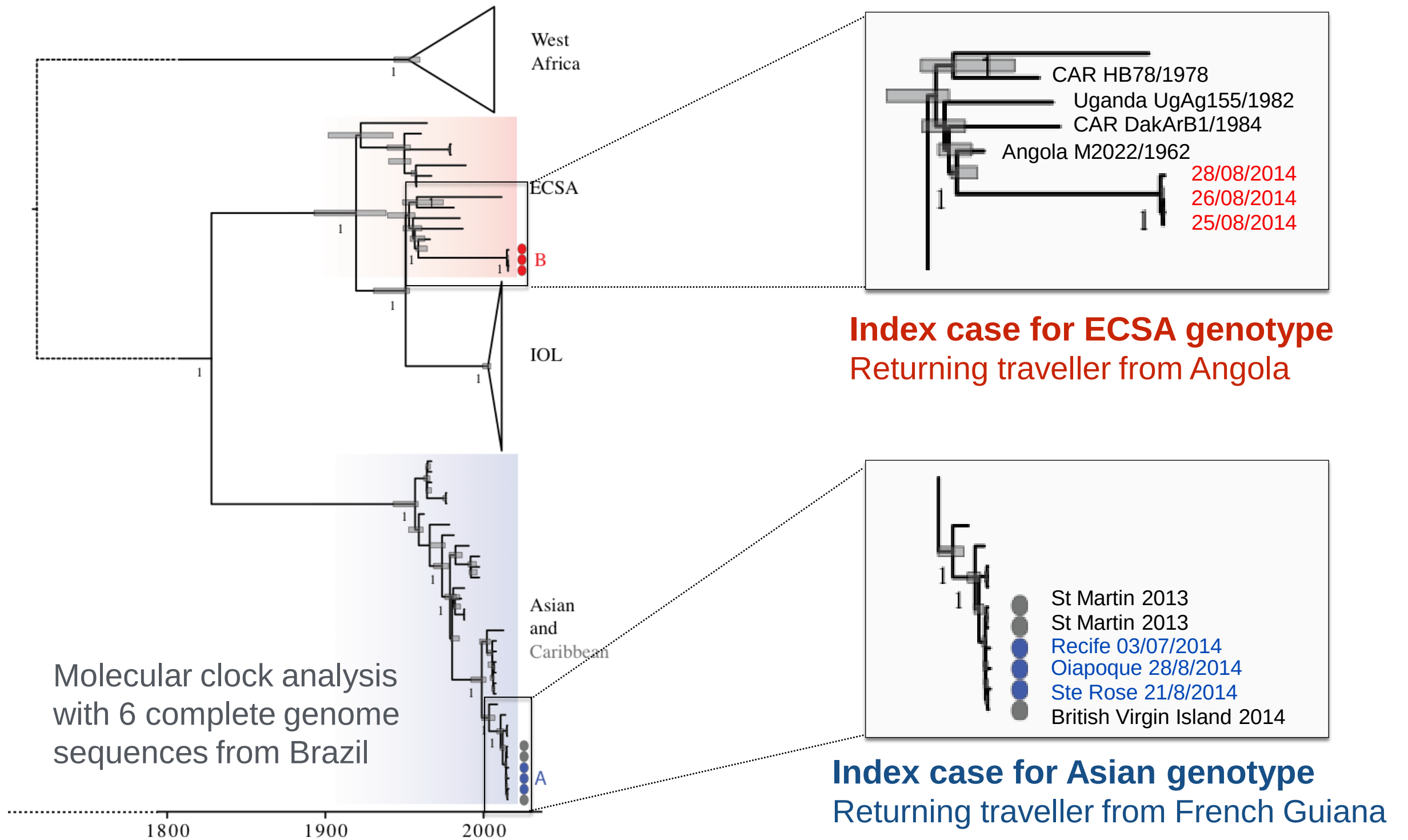


CHIKV-Asian Caribbean:
23 Aug 2012 (3 Oct 2011 - 02 Jun 2013)

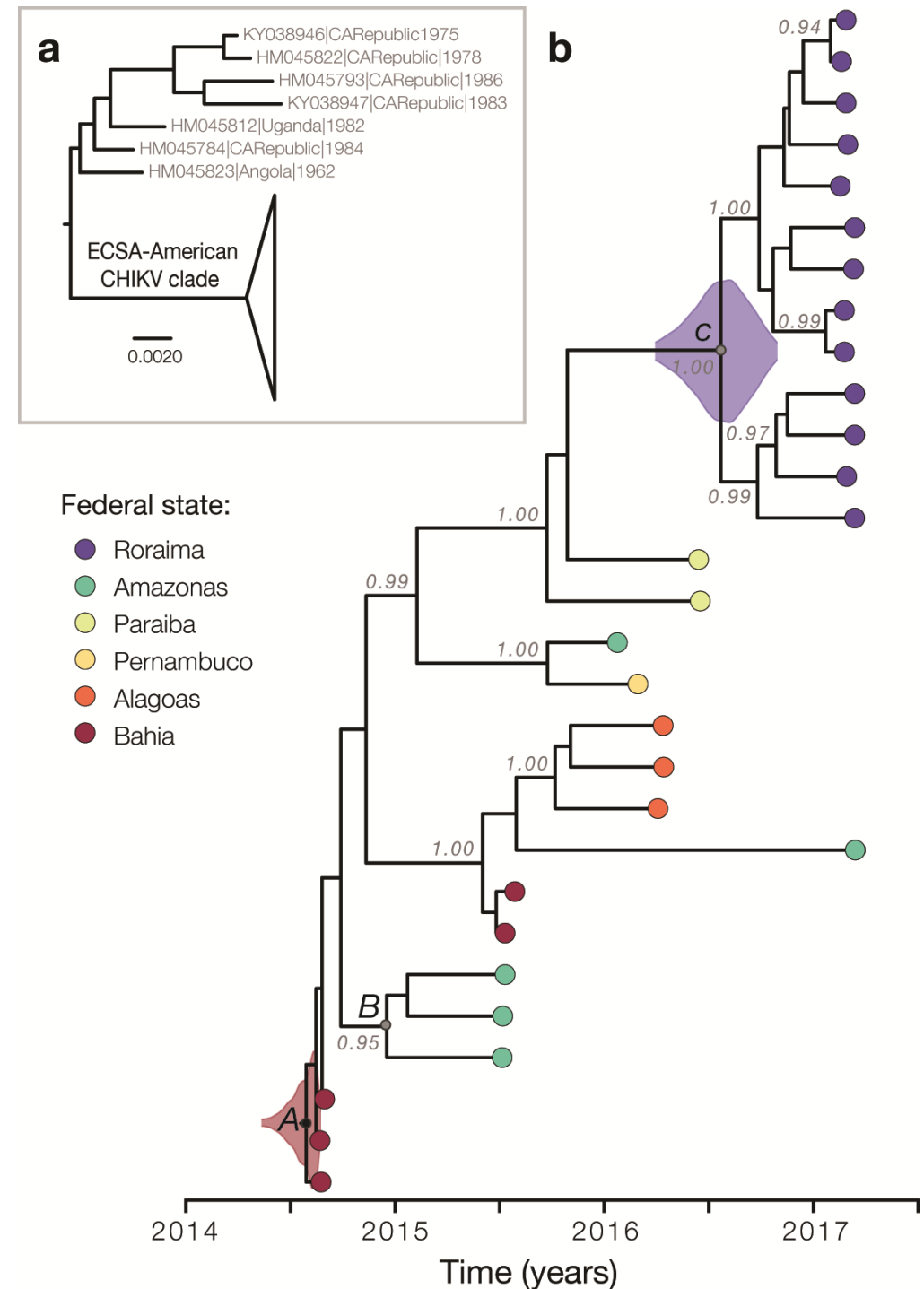
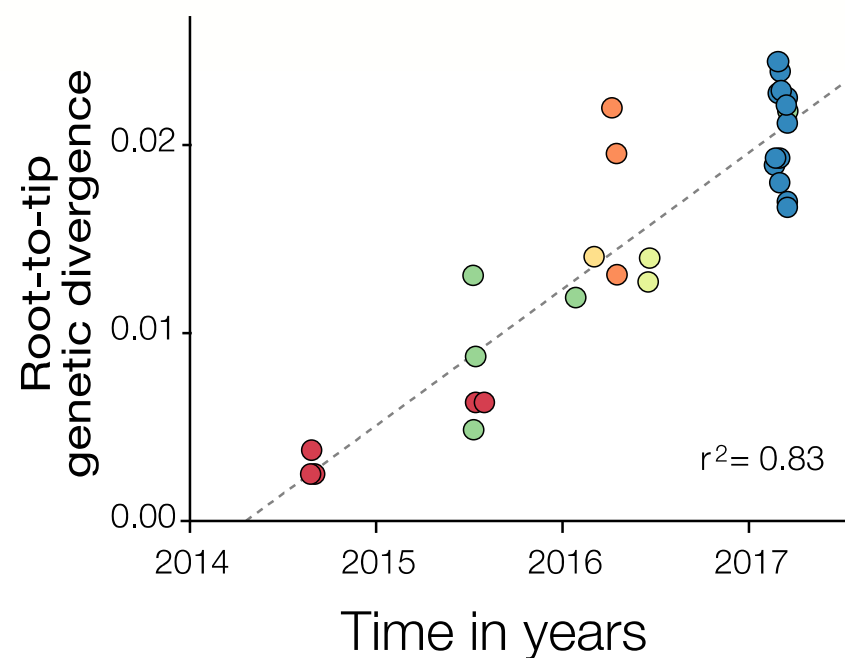
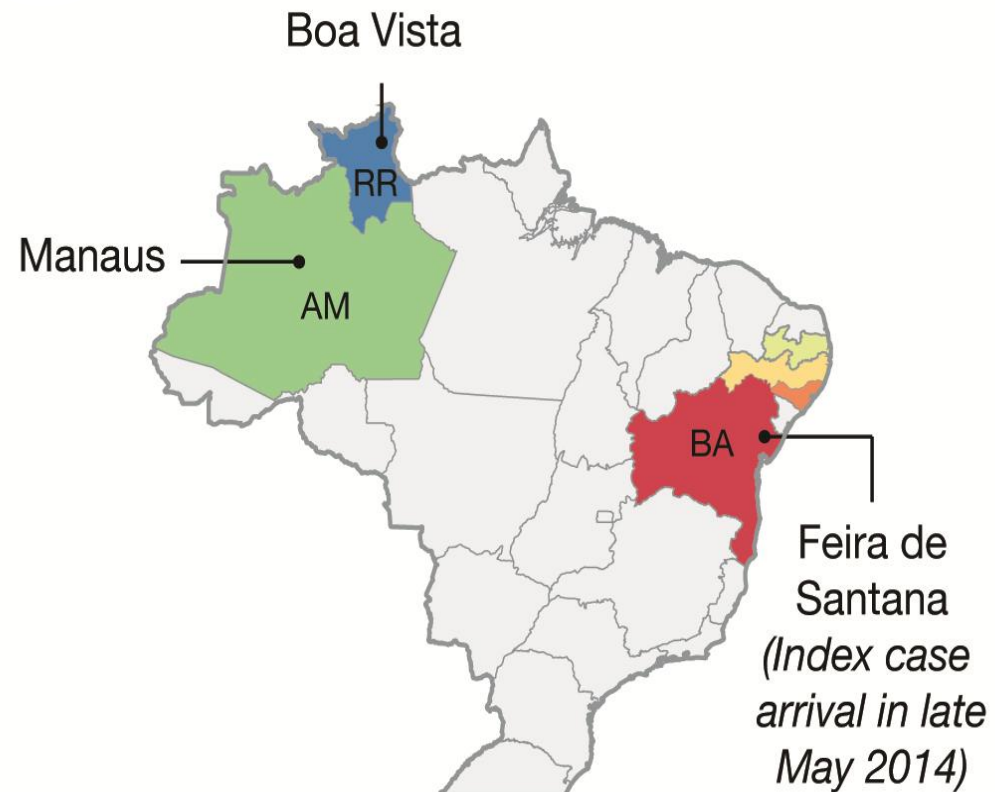
CHIKV-ECSA Feira de Santana:
6 Mar 2014 (29 Jul 2013 - 15 Aug 2014)

Distinct CHIKV genotypes circulating in Brazil

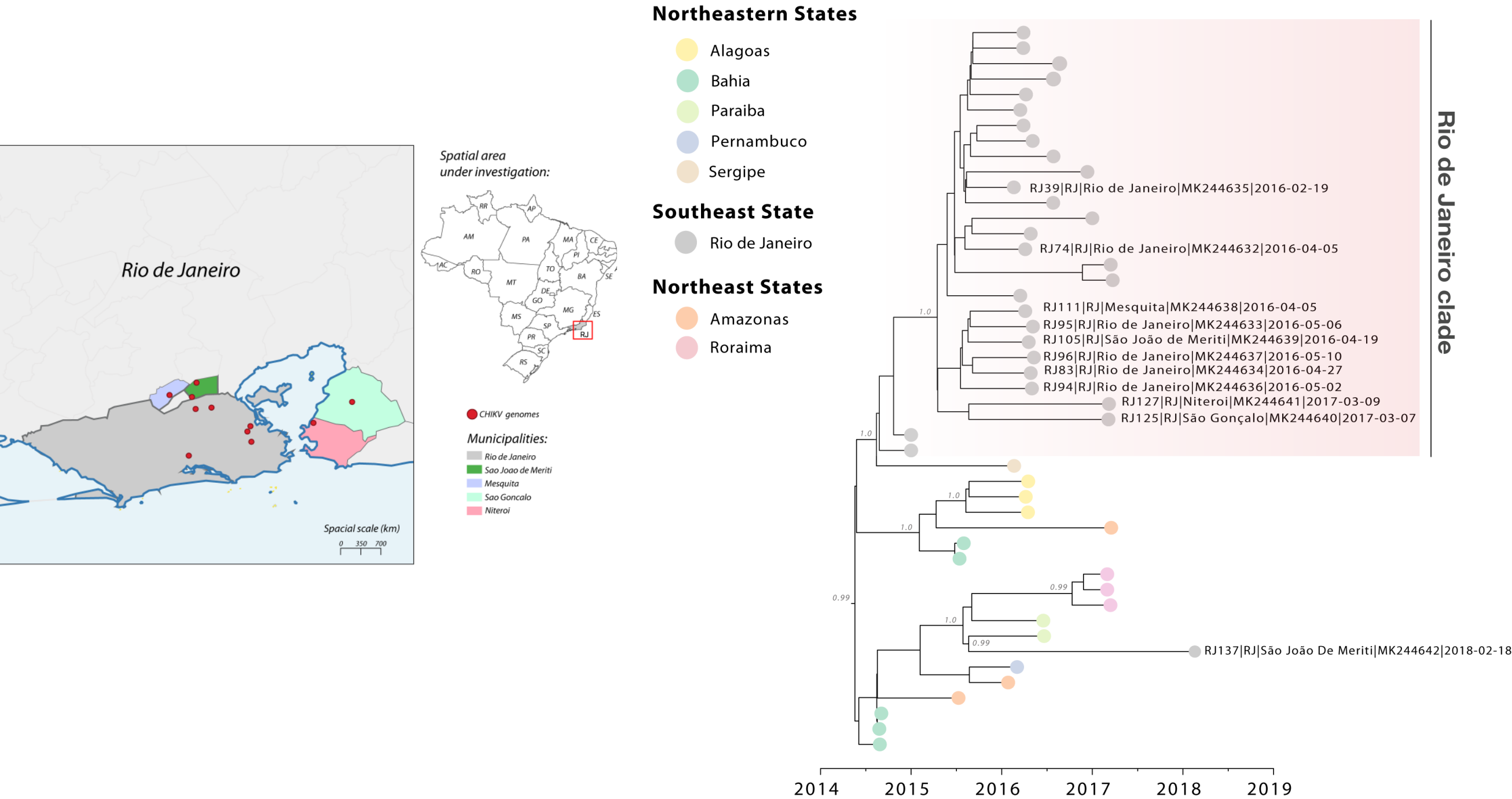
CHIKV-ECSA introduced in Brazil from Angola



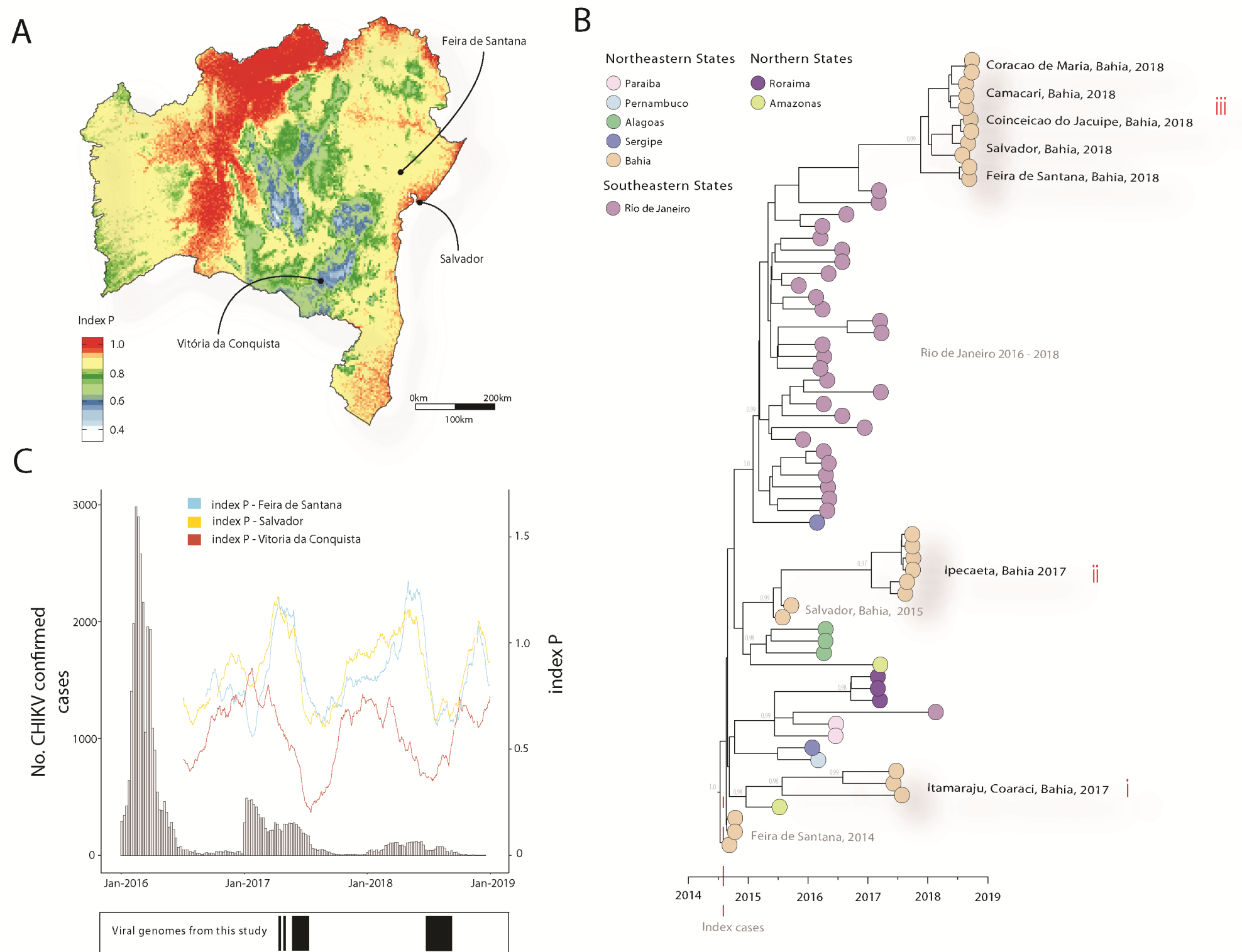
Surto do vírus Chikungunya no Norte do Brasil: substituição do genótipo asiático para o ECSA?



Circulation of chikungunya virus East/Central/South African lineage in Rio de Janeiro, Brazil



Return of the founder Chikungunya virus to its place of introduction into Brazil is revealed by genomic characterization of exanthematic disease cases



Parte 1

Vigilância Genômica do vírus ZIKV

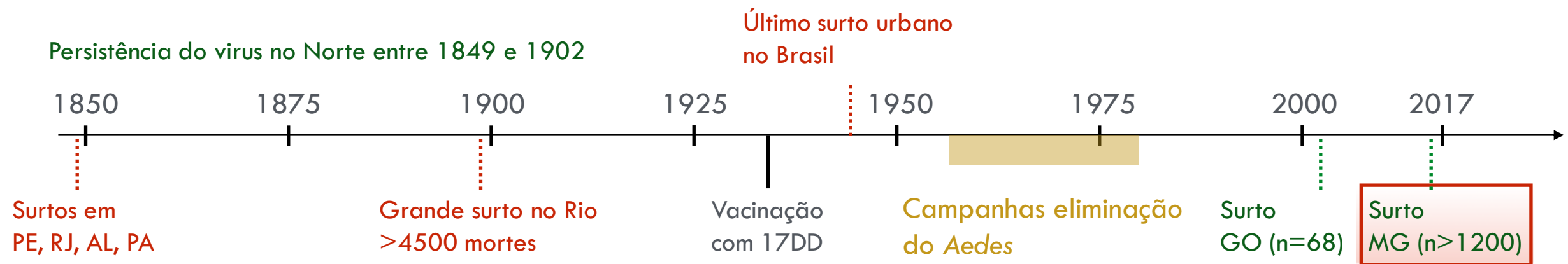
Parte 2

Vigilância Genômica do vírus CHIKV

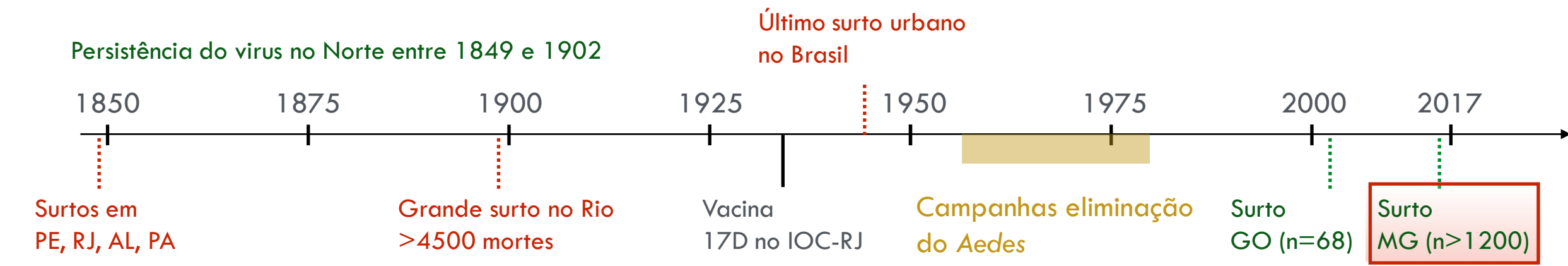
Parte 3

Vigilância Genômica do vírus YFV

História da febre amarela no Brasil

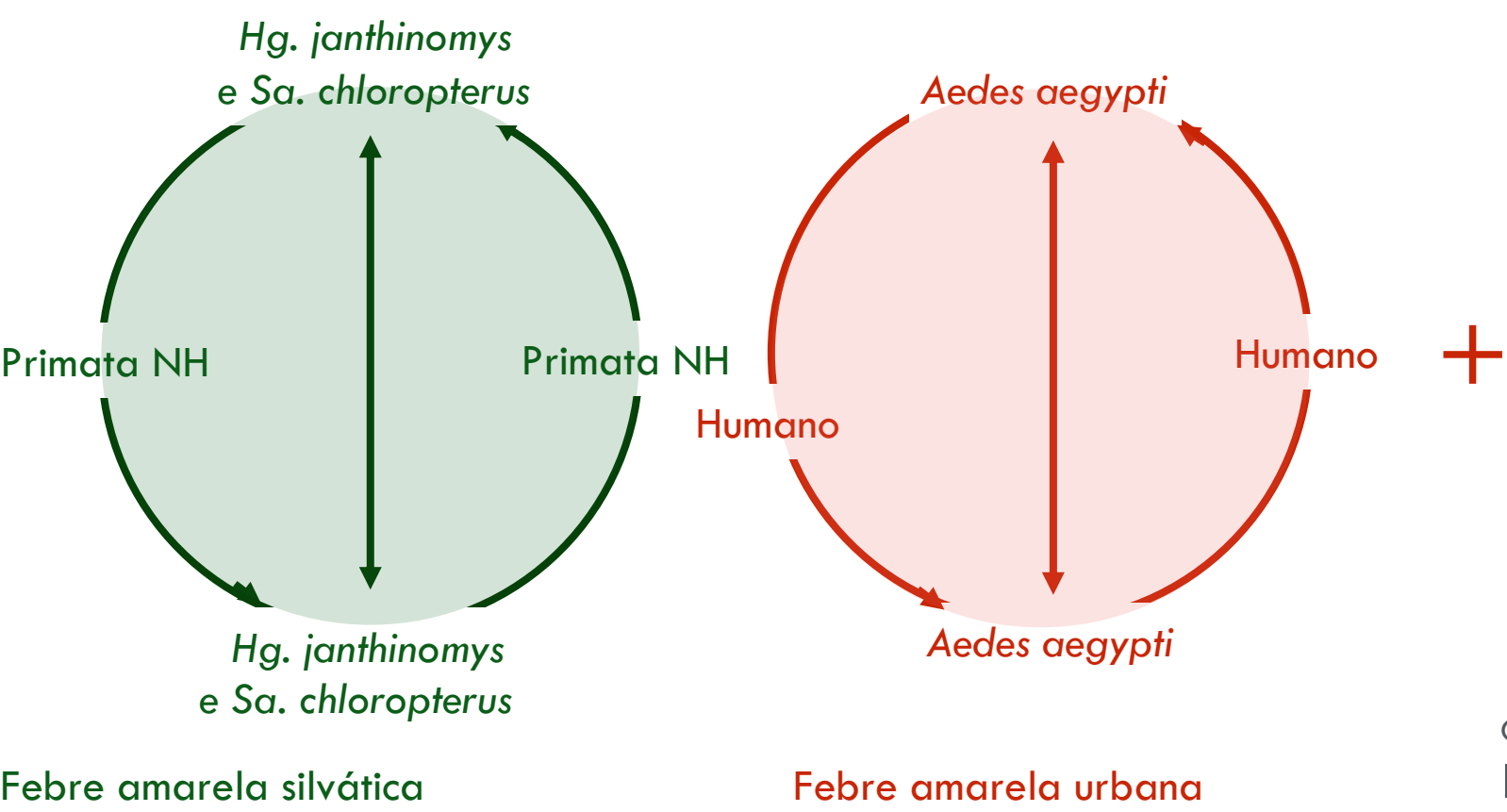


História da febre amarela no Brasil

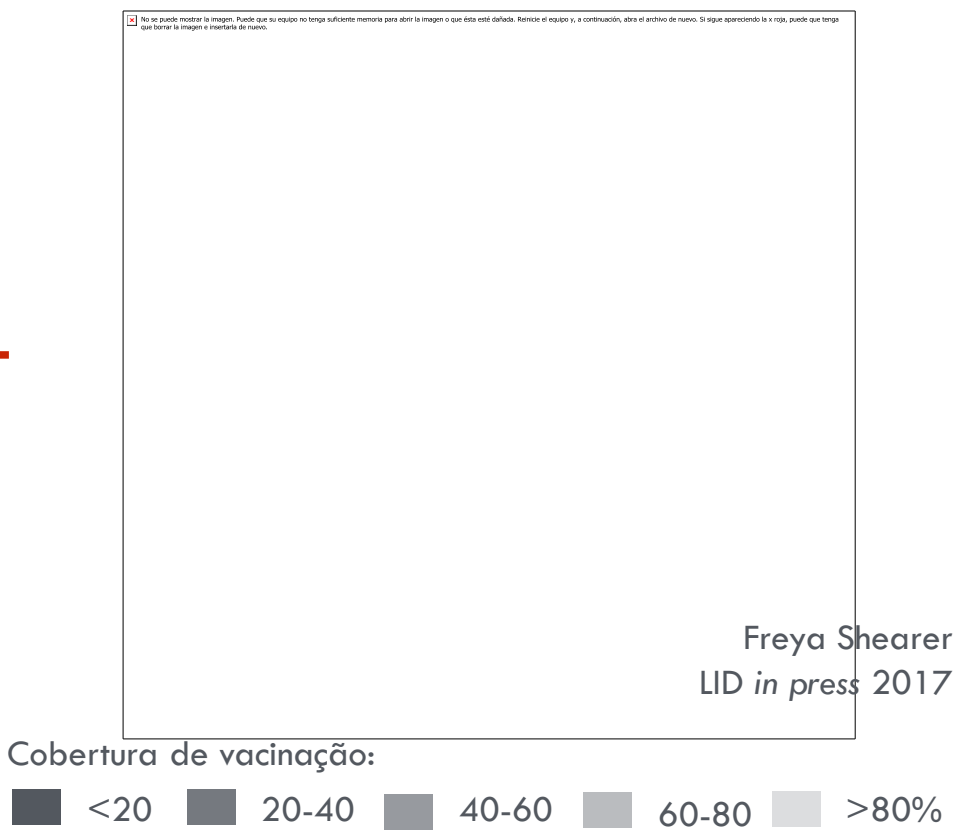


Vainio & Cutts, WHO 1981

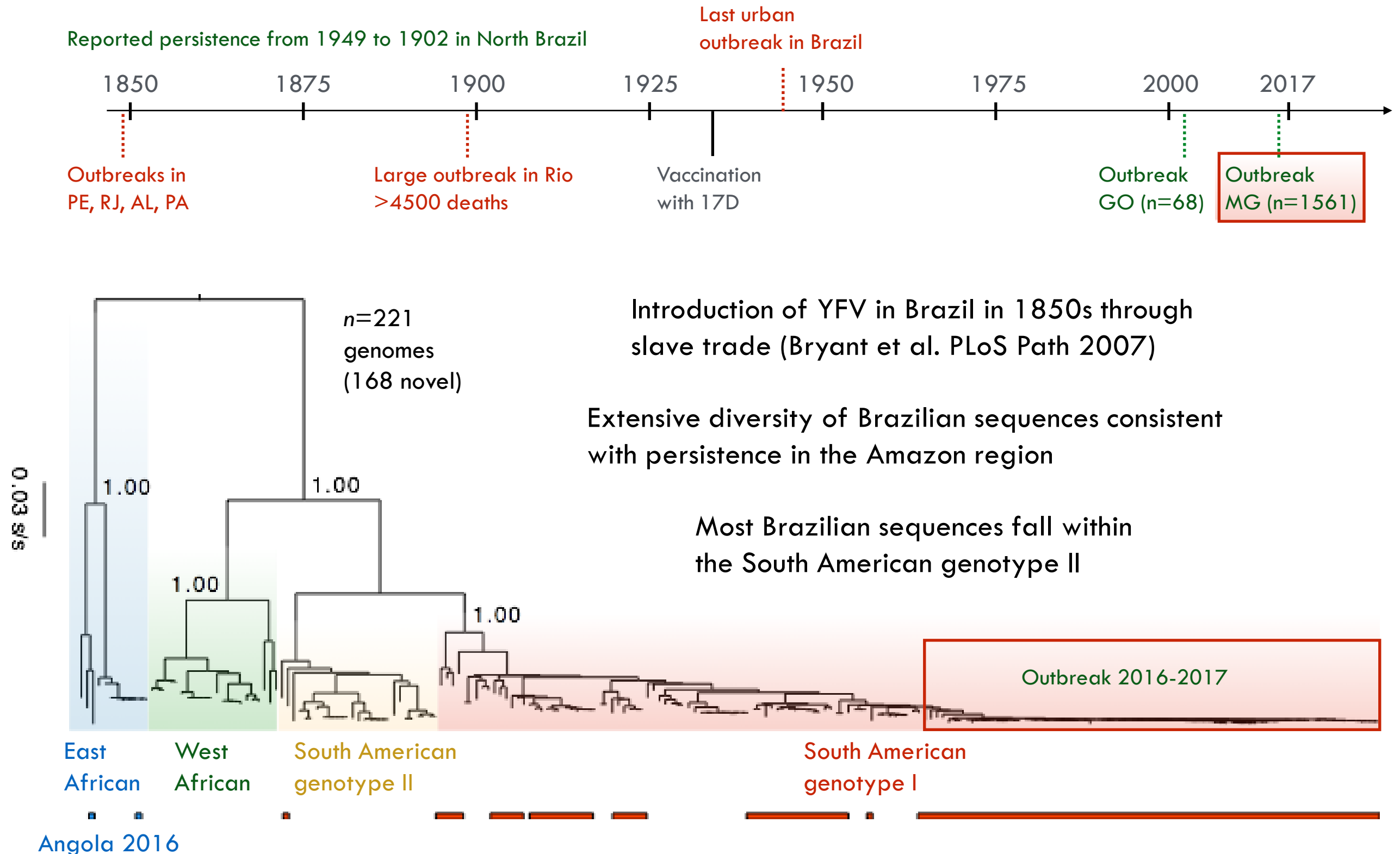
Ciclo de transmissão na América do Sul



Cobertura vacinação contra febre amarela no Brasil 2016

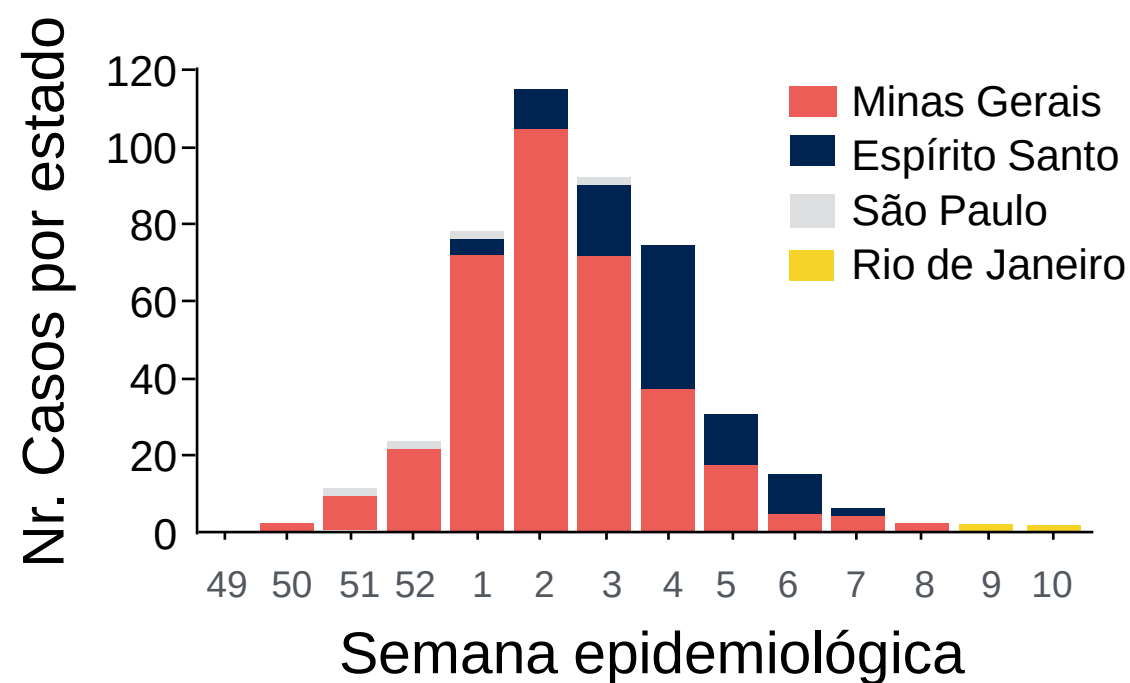
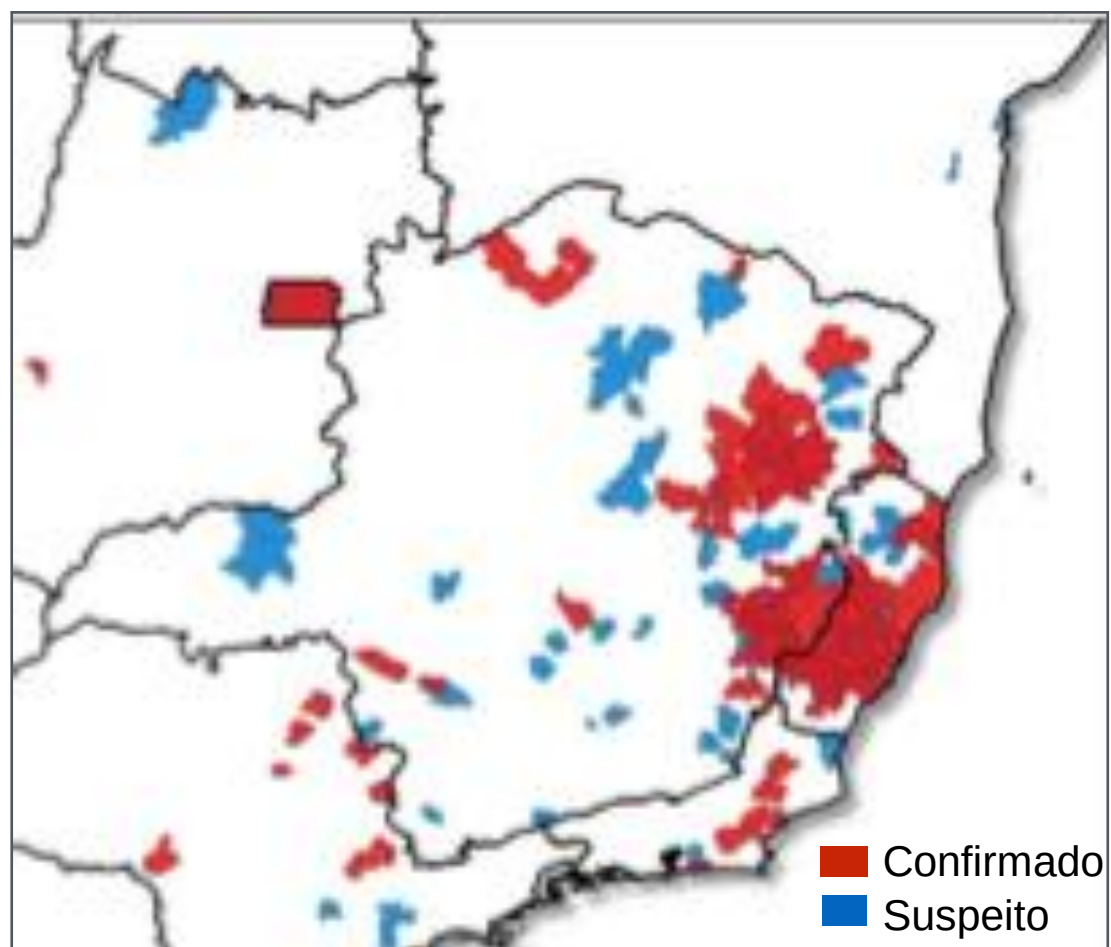


História da febre amarela no Brasil

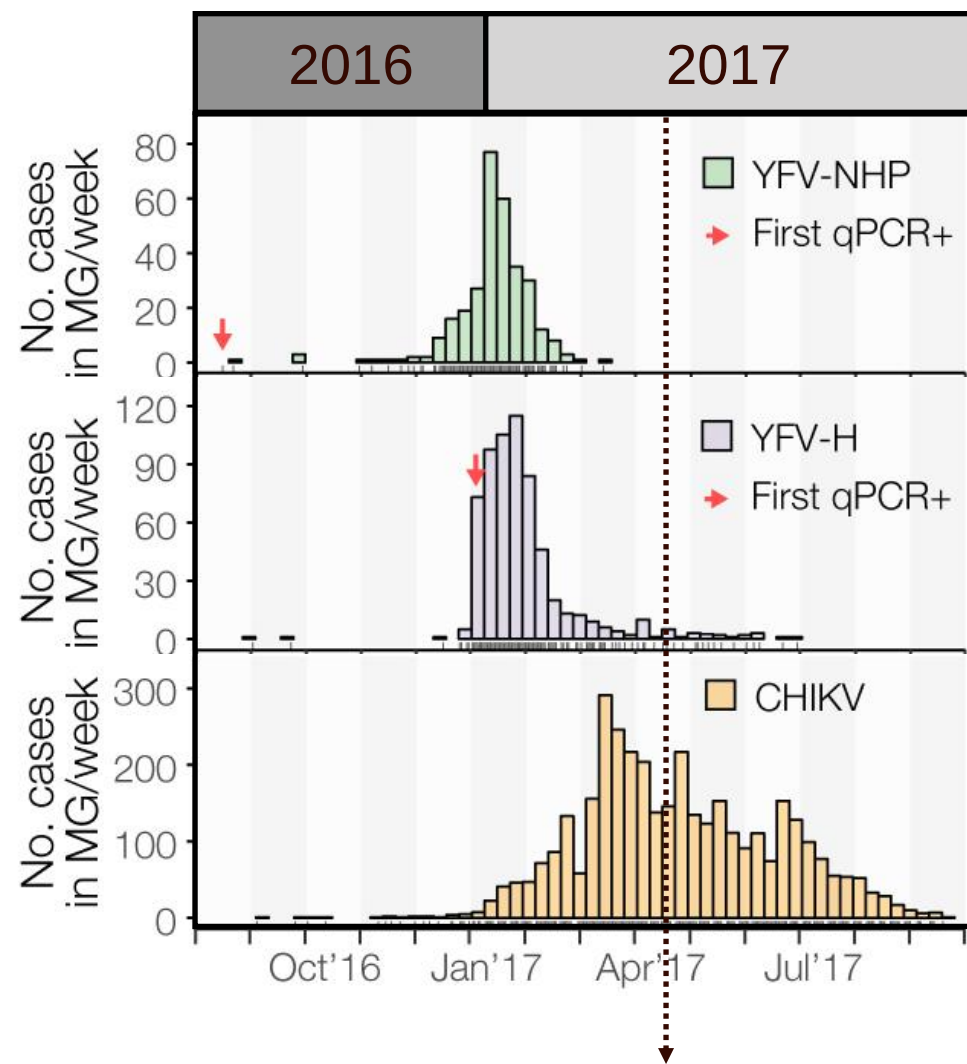


Surto de febre amarela em Minas Gerais

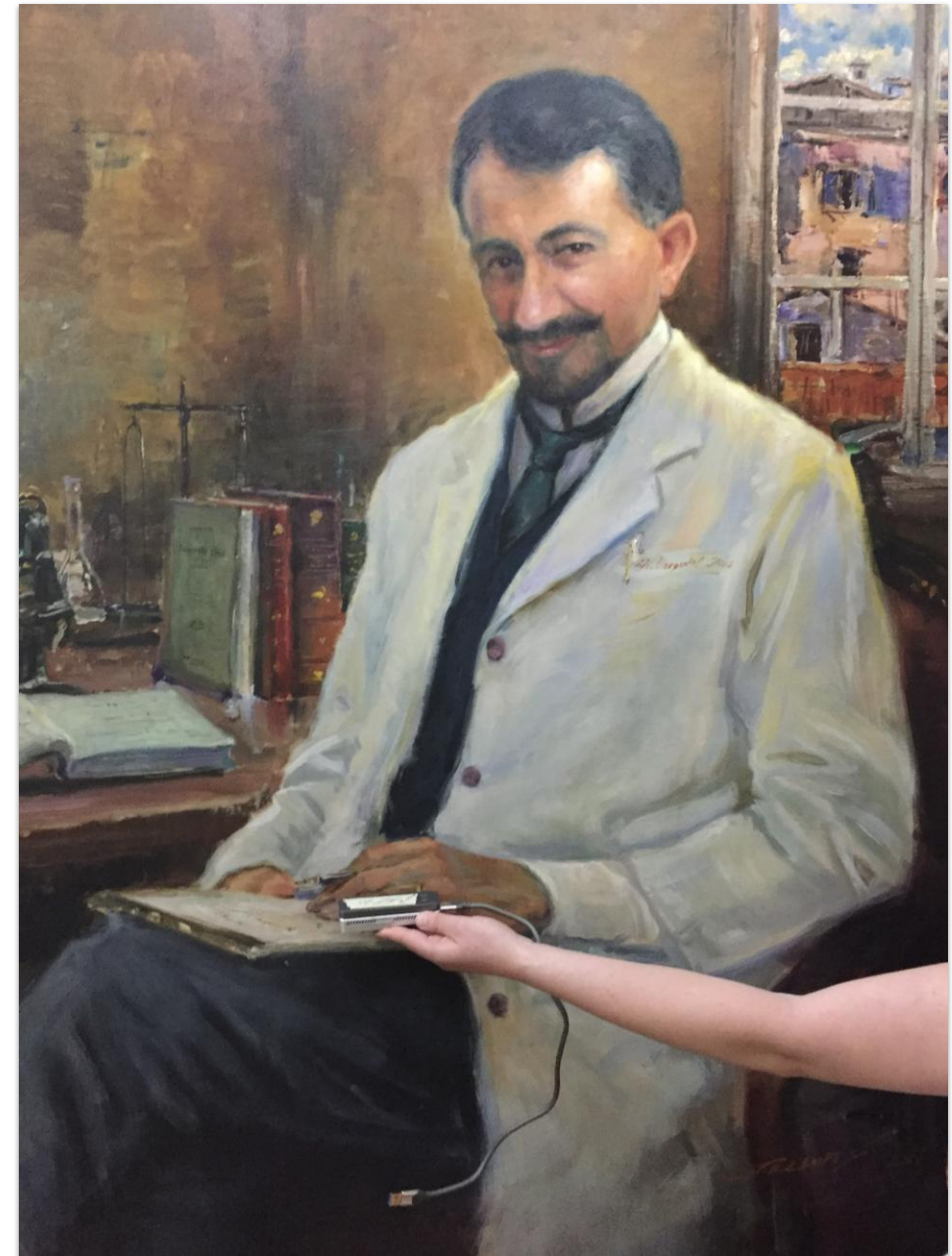
Dez 2016: primeiros casos confirmados em MG
31% taxa de fatalidade por caso (ECDC 2017)
Abril 2017: 709 casos, >250 mortes
Ciclo silvático (ou transmissão urbana?)



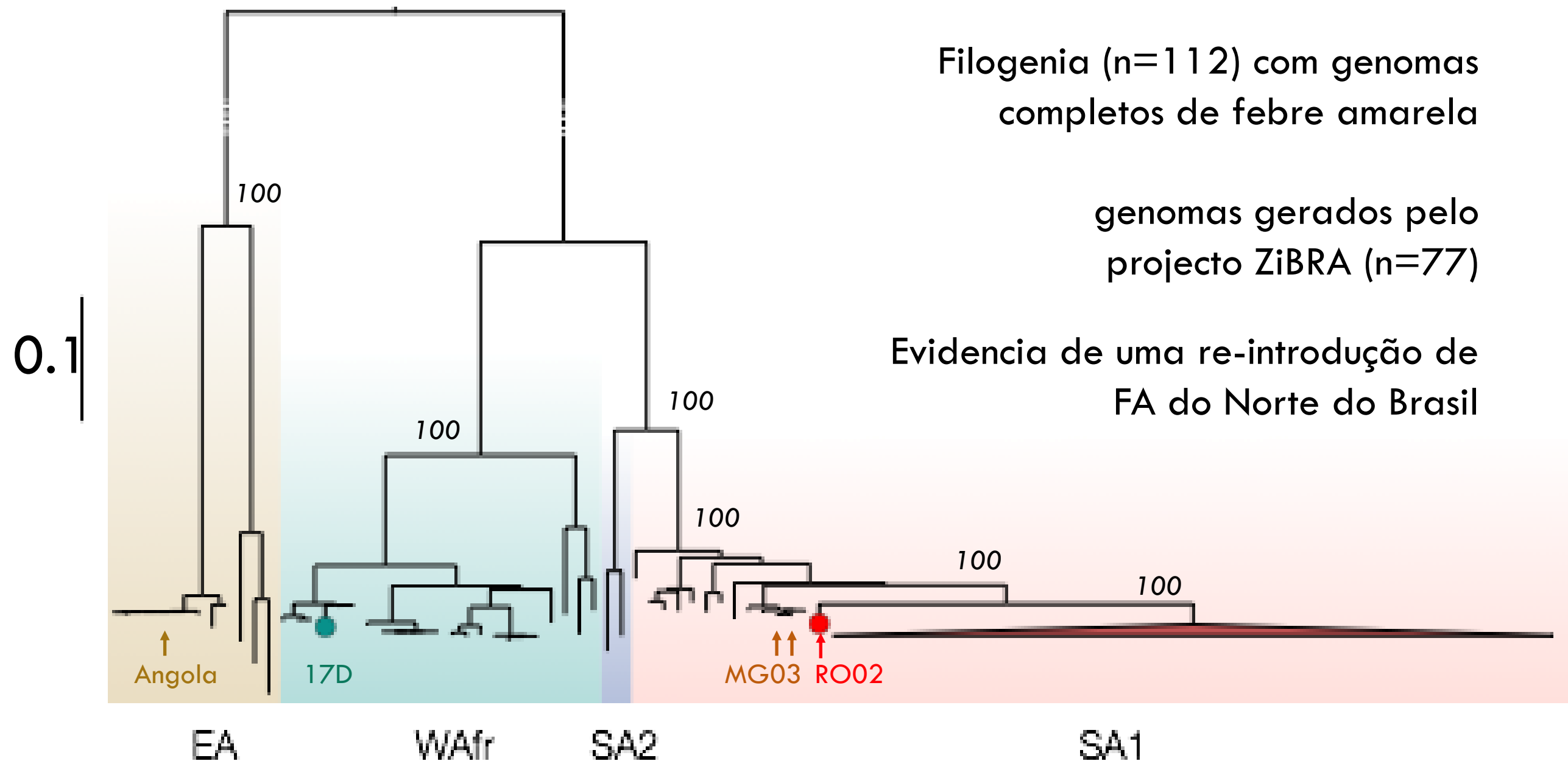
Associação espacial e temporal de casos de primatas humanos e não humanos



Vigilância genômica do YFV
realizada em colaboração com o
LACEN-FUNED

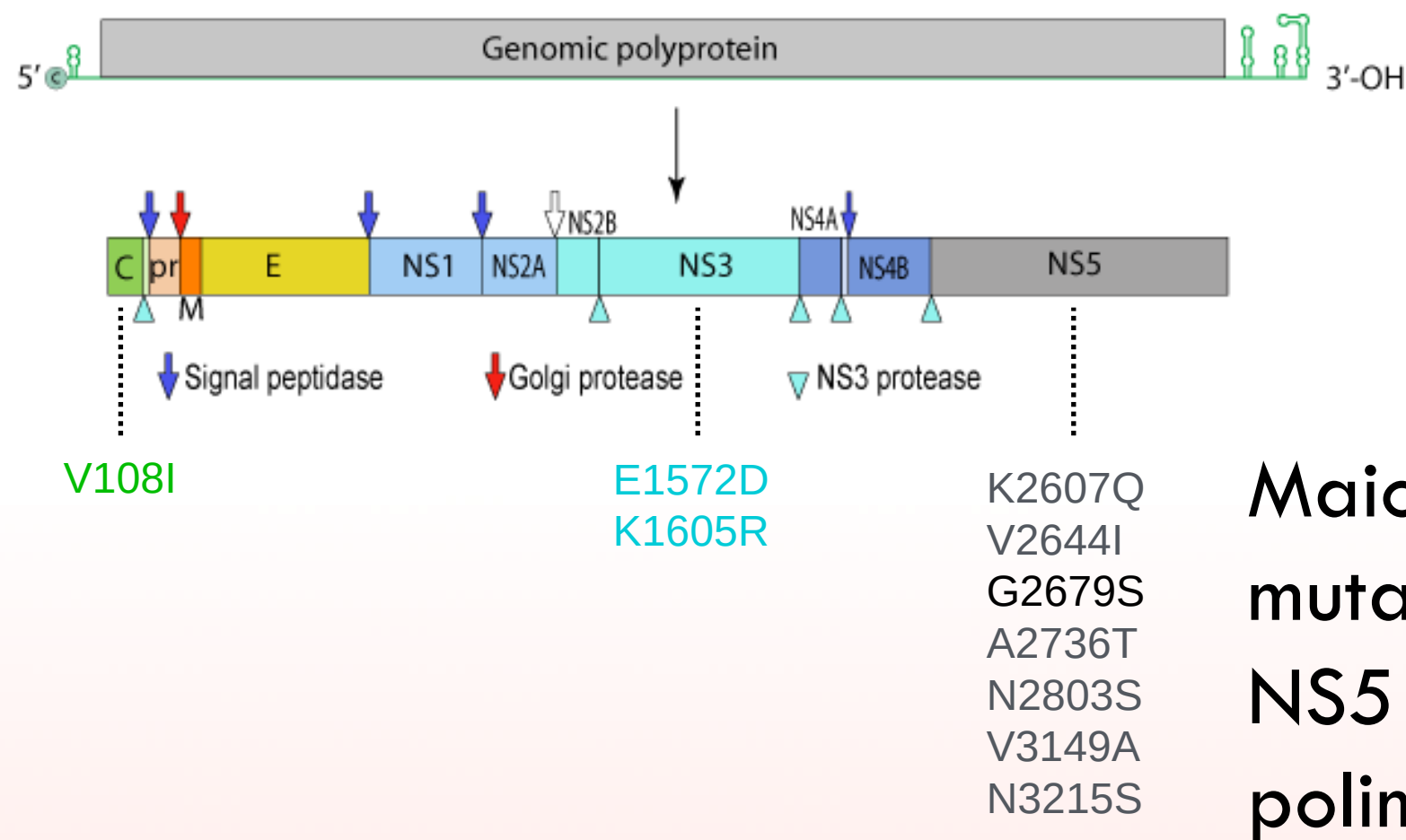


Epidemiologia genómica do surto de febre amarela no sudeste Brasileiro

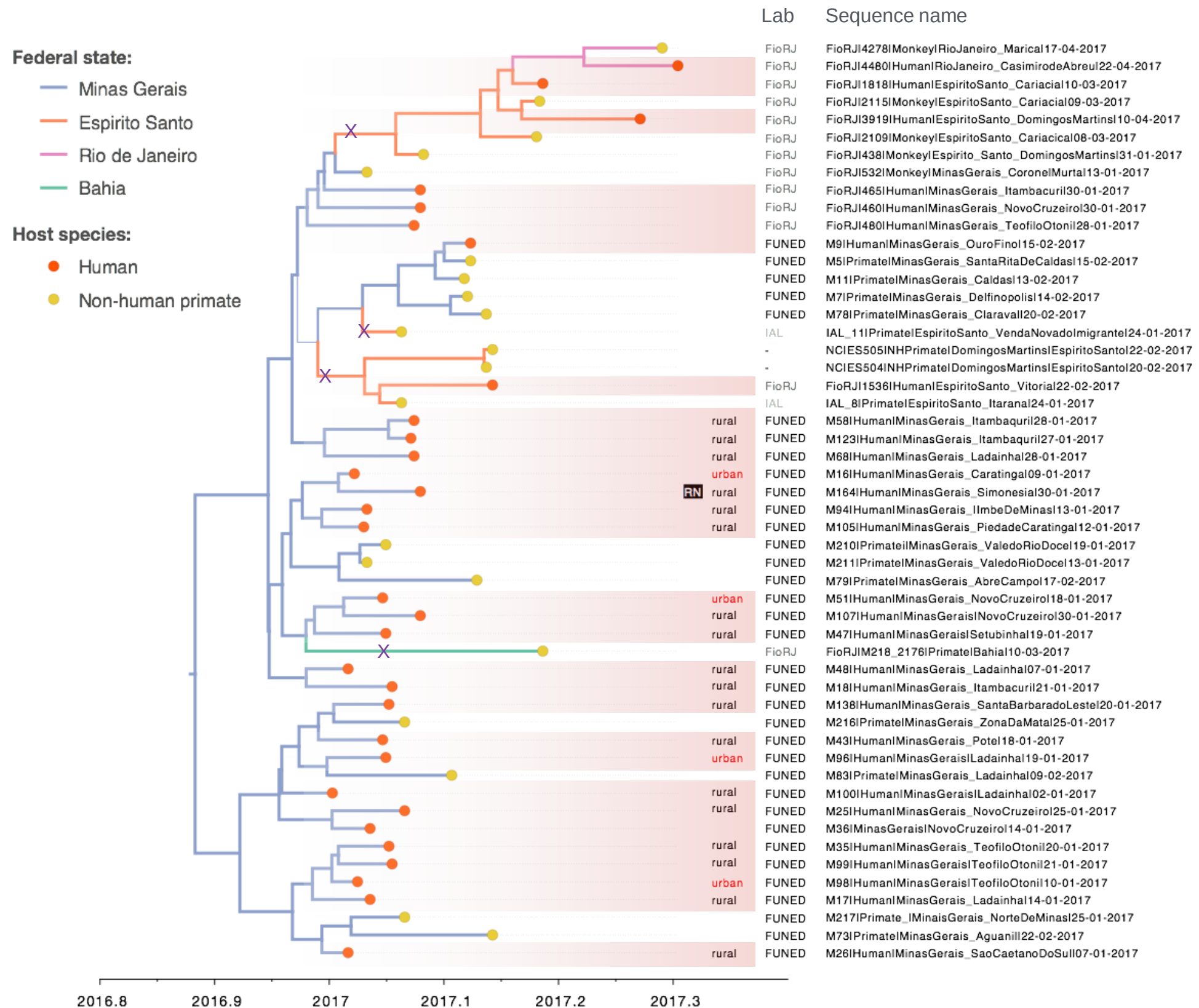


Determinantes genéticos do surto de febre amarela no sudeste Brasileiro

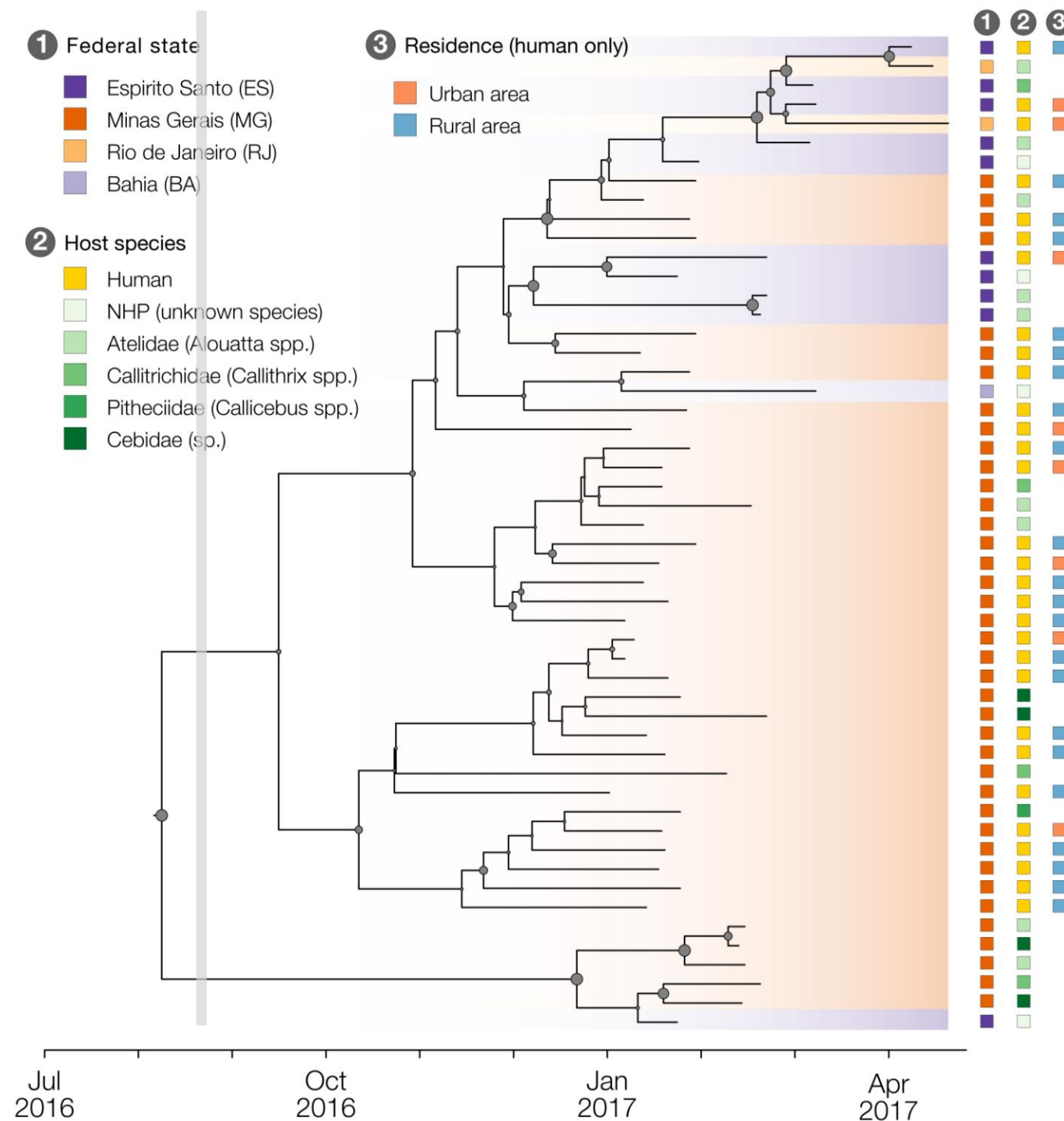
****** mutações únicas do surto 2016-2017



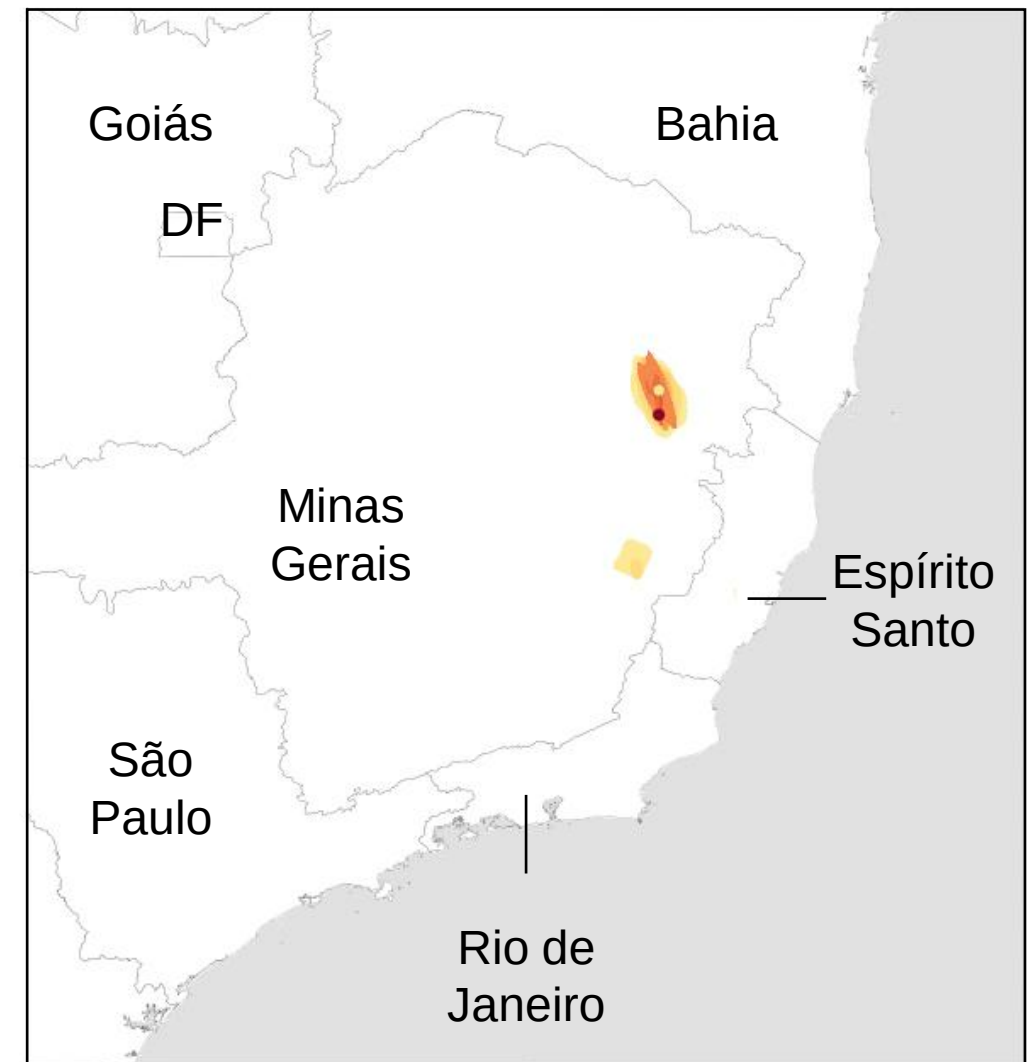
Origens e evolução do surto de FA em MG



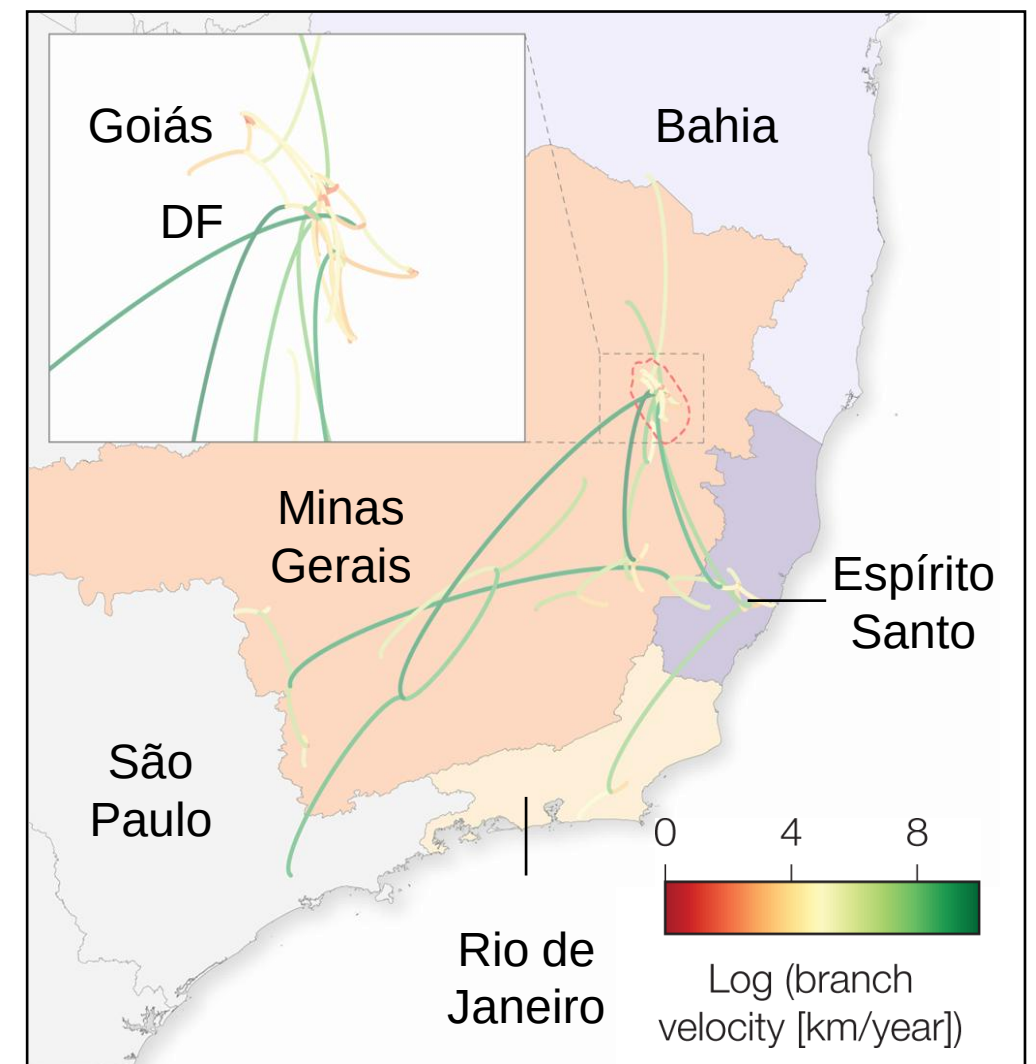
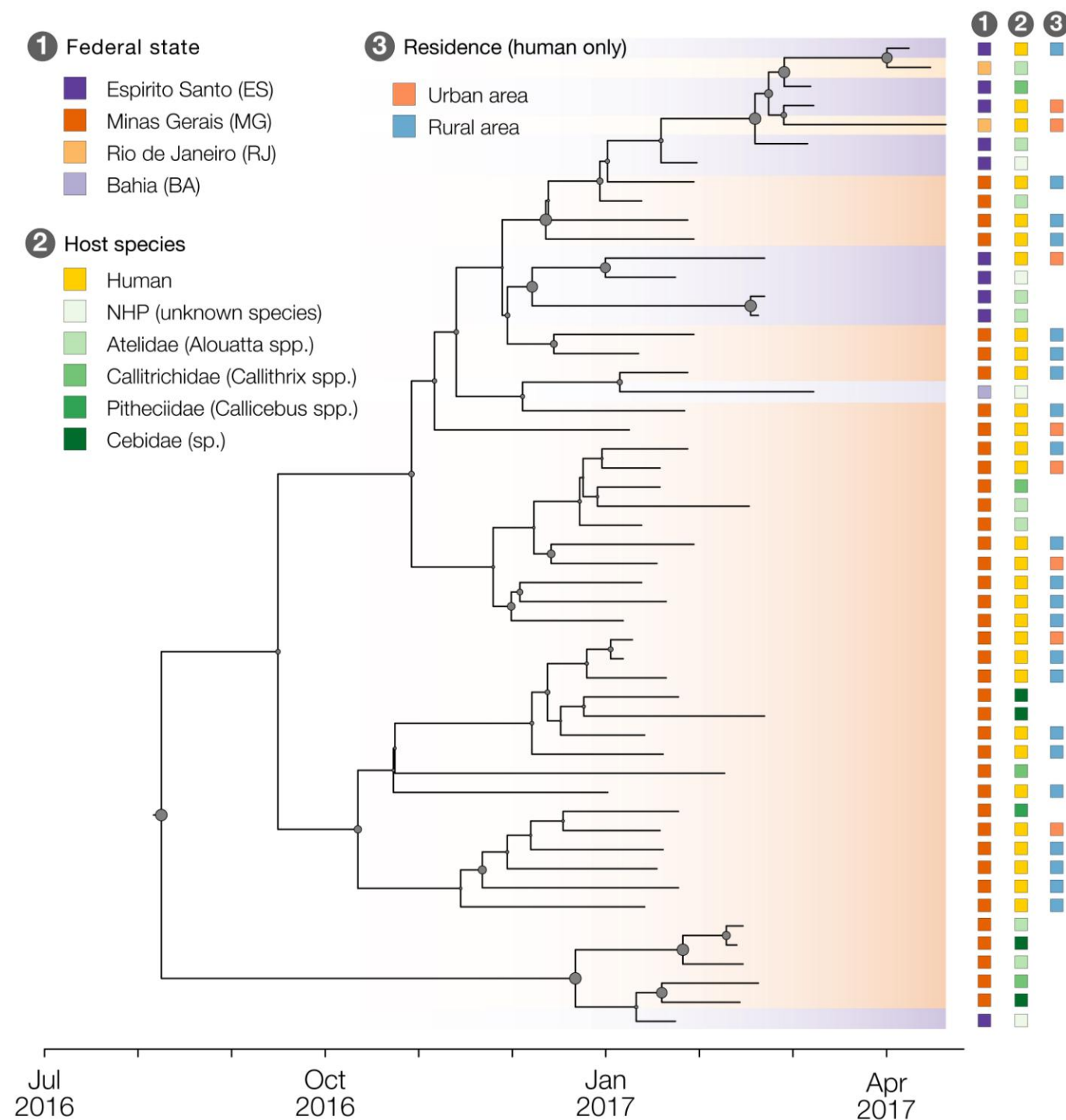
Surto de YFV surgiu em agosto de 2016 e se espalhou várias vezes para outros estados



Dispersão de FA
para o Este da região do Sudeste

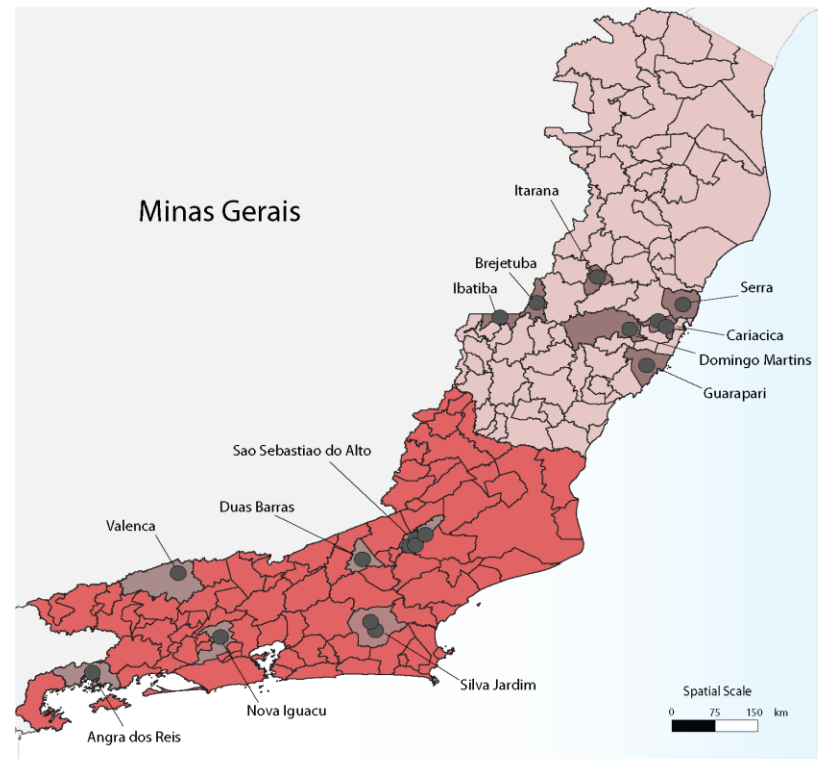


Vários movimentos de longa distância sugerem dispersão do YFV mediada por humanos



Yellow fever virus re-emergence and spread in Southeast Brazil, 2016-2019

A

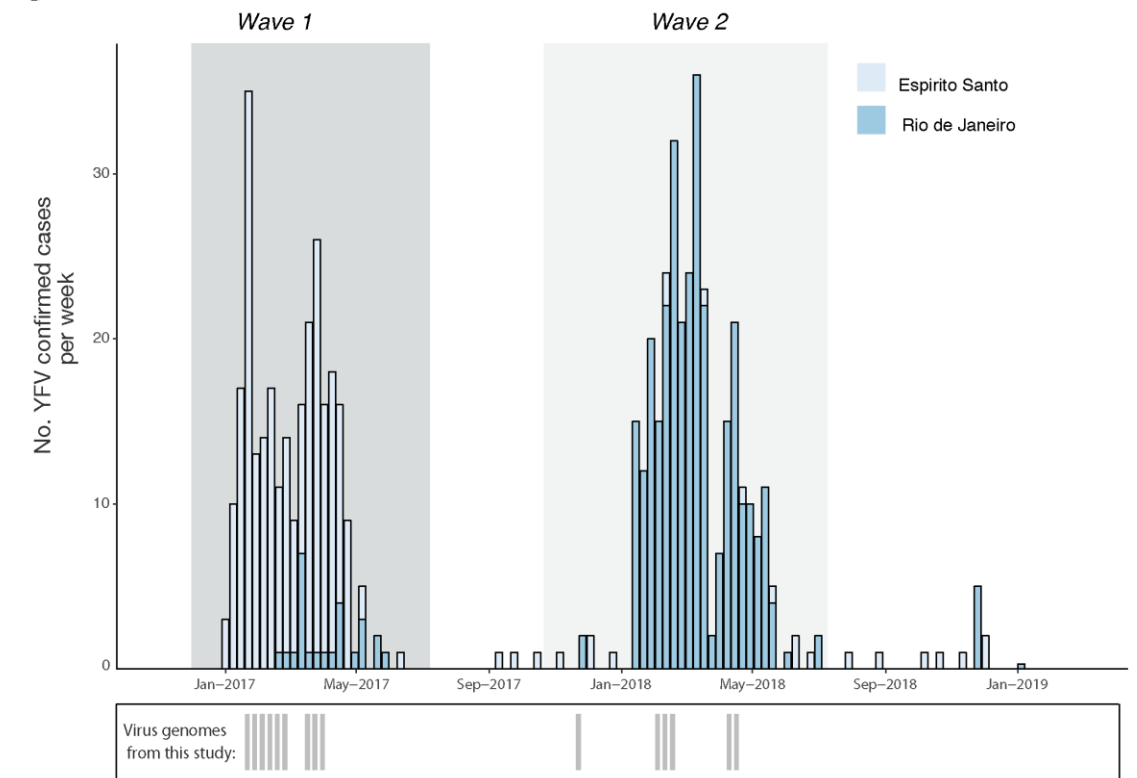


Spatial area under investigation:



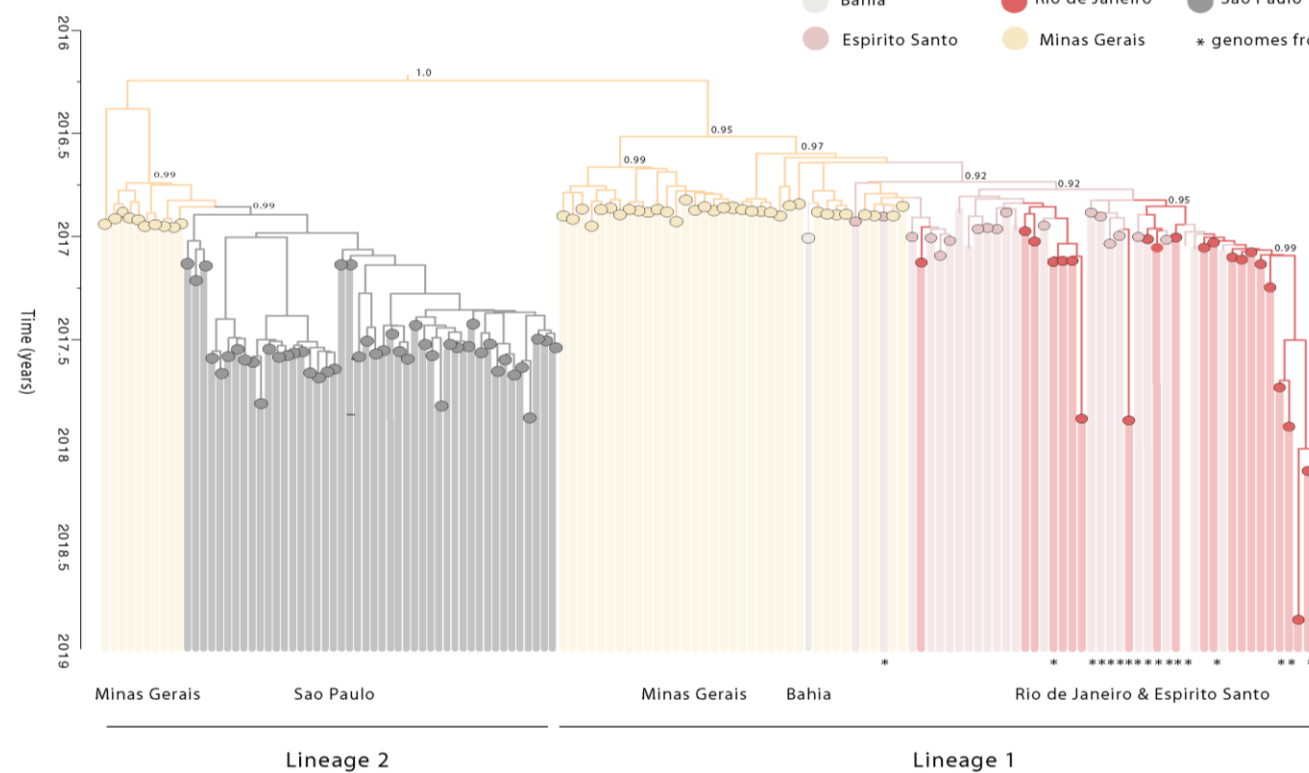
● genomes from this study

B



Sampling Location

Bahia Rio de Janeiro Sao Paulo
Espirito Santo Minas Gerais * genomes from this study

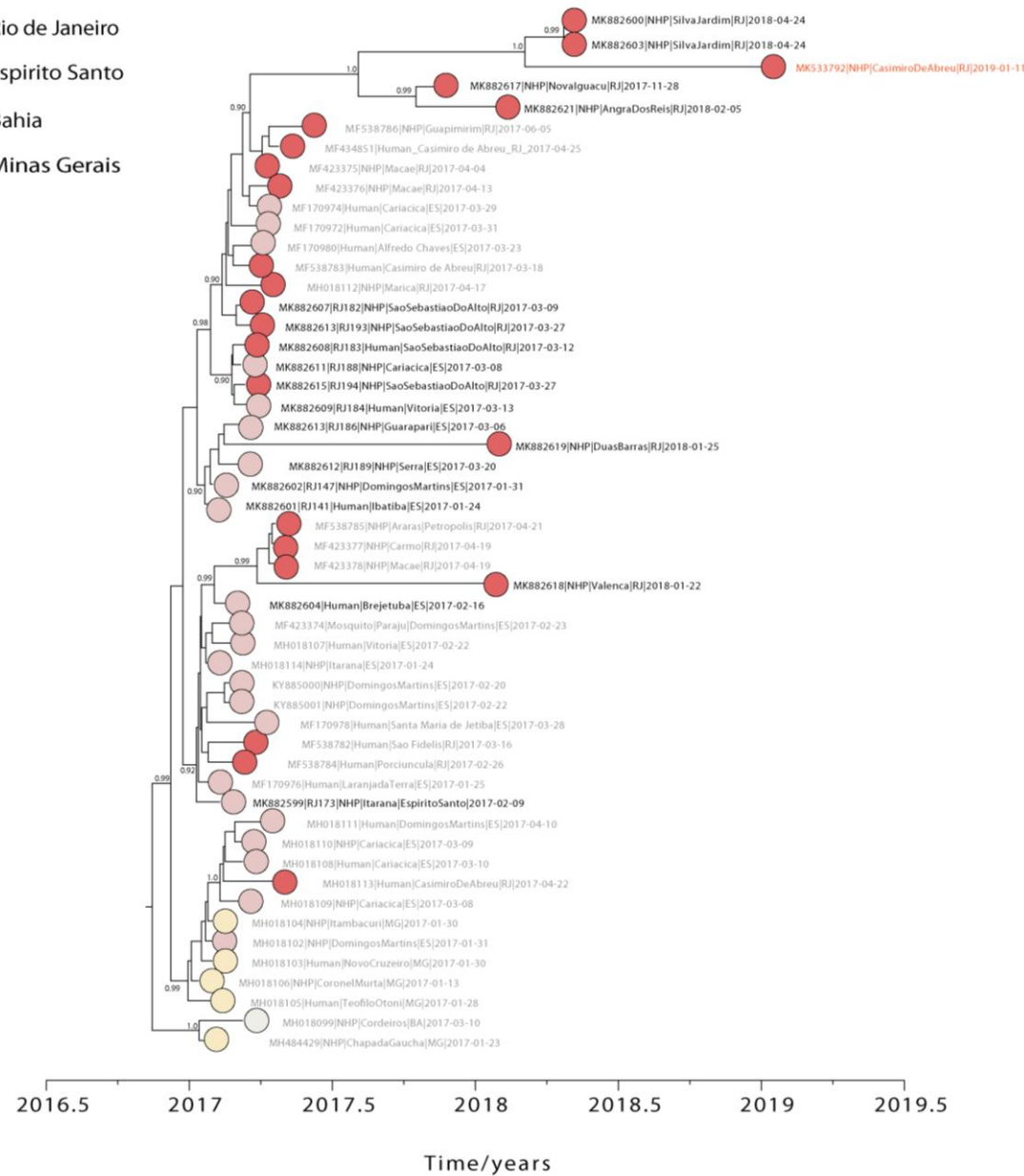


Yellow fever virus re-emergence and spread in Southeast Brazil, 2016-2019

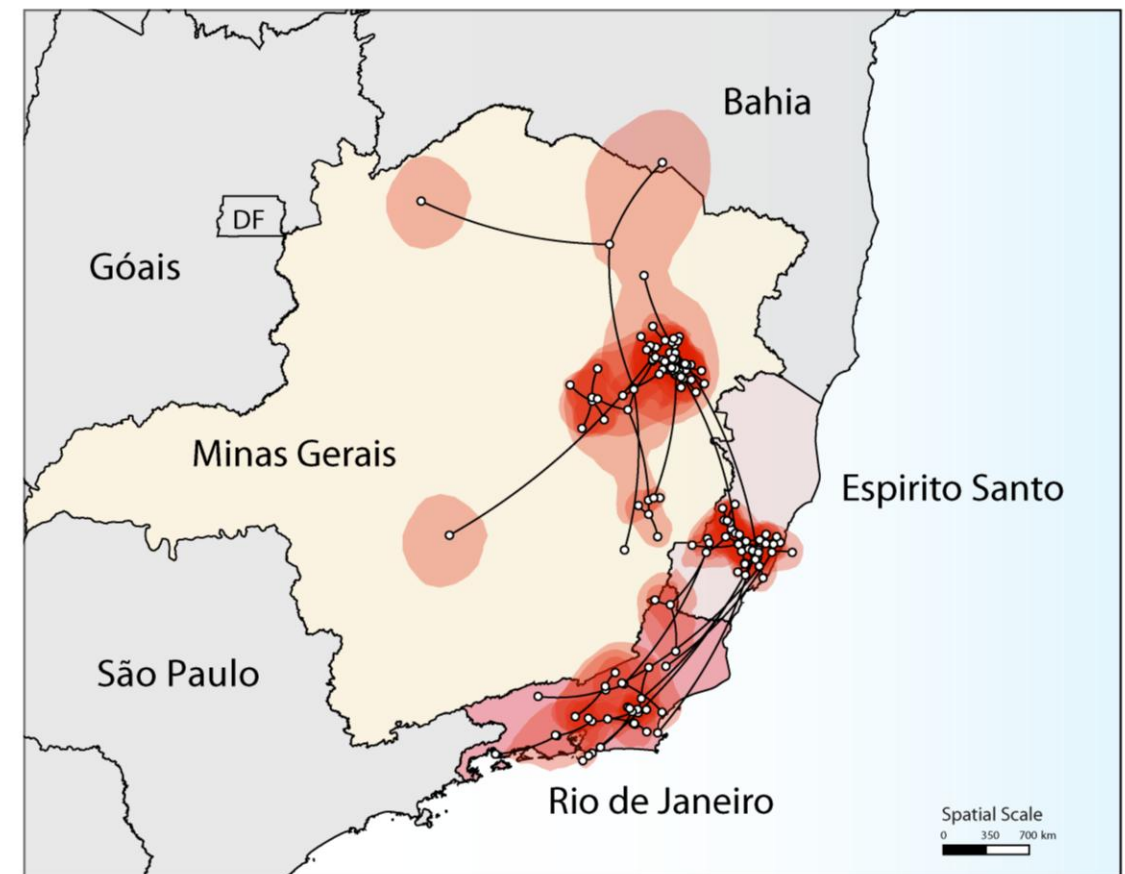
A

Sampling Locations

- Rio de Janeiro
- Espírito Santo
- Bahia
- Minas Gerais



B



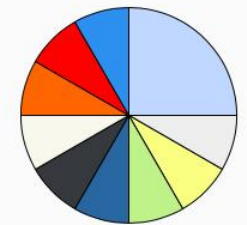
Desenvolvimento de ferramentas



Analysis result

You may bookmark this page to revisit results of this job (541435477) later.

Rega assignment	Sequences count	Percentage	Source	Legend
Zika virus	3	25	NCBI	
Chikungunya virus	1	8.3	NCBI	
Dengue virus 2	1	8.3	NCBI	
Coxsackievirus B3	1	8.3	ICTV	
Human gammaherpesvirus 8	1	8.3	NCBI	
Simian T-lymphotropic virus 1	1	8.3	NCBI	
Yellow fever virus	1	8.3	NCBI	
Norovirus GI1	1	8.3	NCBI	
Hepatitis C virus	1	8.3	ICTV	
Human immunodeficiency virus 1	1	8.3	NCBI	
Totals	12	100		



Download results: [Table \(Excel format\)](#) [Table \(CSV format\)](#) [Sequences \(Fasta format\)](#)



DENGUE, ZIKA & CHIKUNGUNYA VIRUSES TYPING TOOL

Zika Virus Typing Tool Version 0.9 - Alpha

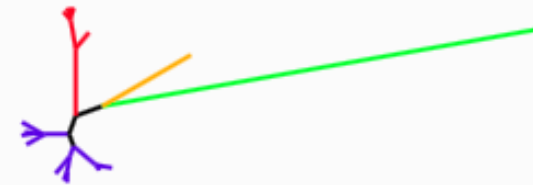
[Submit Job](#) [Monitor job \[272801805\]](#) [How to cite](#) [Introduction](#) [How to use](#) [Example sequences](#)

Zika Virus Typing Tool Results

You may bookmark this page to revisit results of this job (272801805) later.

A whole genome typing will take approximately 60 seconds, please be patient.

[zikv](#)



Name	Length	Virus	Serotype/Clade	Report	Genome
KR815989_Brazil_2015_zika	330	Zika Virus		Report	
KU955594_Uganda_1947	630	Zika Virus		Report	
KF383120_2001	700	Zika Virus		Report	

Download results: [XML File](#) [Table \(Excel format\)](#) [Table \(CSV format\)](#) [Sequences \(Fasta format\)](#)

Developed by: **FIOCRUZ/Bahia, Brazil** (Maria Inés Restovic, Marta Giovanetti, Vagner Fonseca, Murilo Freire, Luiz Alcantara), **KU Leuven, Belgium** (Kristof Theys, Pieter Libin, Lize Cuypers, Ana Abecasis, Anne-Mieke Vandamme), **Oxford, U.K.** (Nuno Faria and Oliver Pybus), **Evandro Chagas Institute, Brazil** (Marcio Roberto Teixeira Nunes), **CDC/OID/NCEZID** (Gilberto A. Santiago), **Emweb bvba, Belgium** (Koen Deforche) and **Africa Centre/UKZN, South Africa** (Tulio de Oliveira).

Contact: [Dr. Luiz Carlos Junior Alcantara](#), [Dr. Marcio Roberto Teixeira Nunes](#), [Dr. Nuno Faria](#) and/or [Prof. Tulio de Oliveira](#)





DENGUE, ZIKA & CHIKUNGUNYA VIRUSES TYPING TOOL

Zika Virus Typing Tool Version 0.9 - Alpha

[Submit Job](#) [Monitor job \[816727275\]](#) [How to cite](#) [Introduction](#) [How to use](#) [Example sequences](#)

Phylogenetic analysis of all input sequences in zikv.

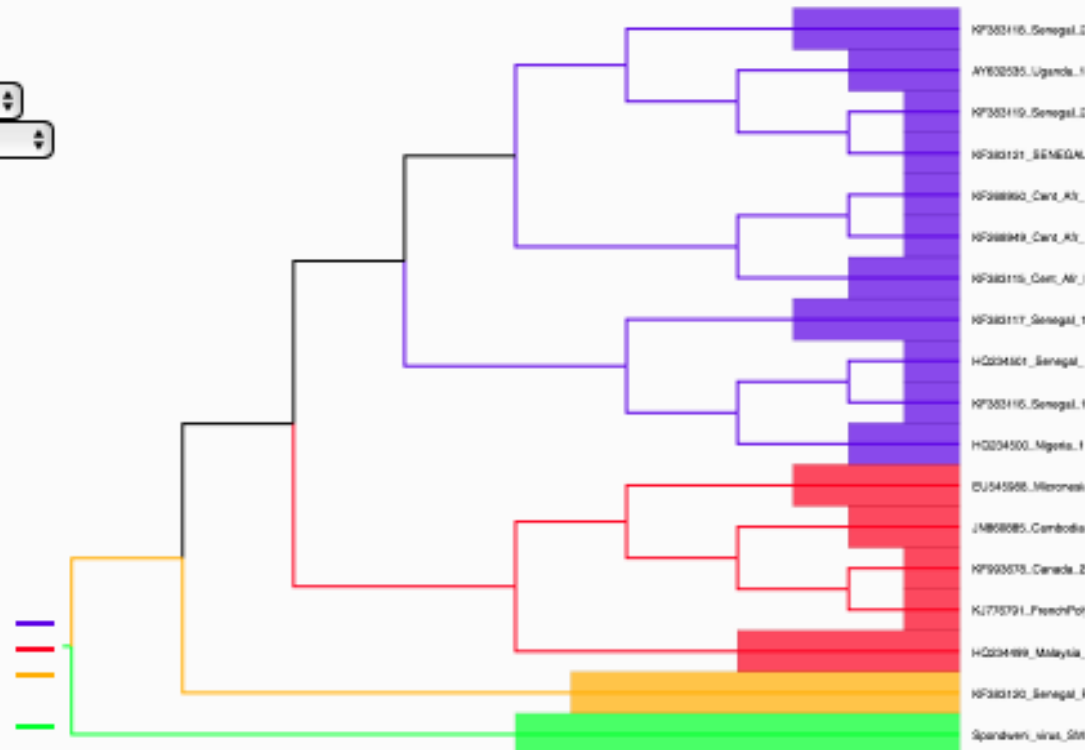
This is a general overview on the input. For a more accurate analysis of a sequence, click on the job tab.

Tree controls

Layout

☒ Show labels
☒ Highlight clusters
☒ Color branches

African
Asian
Divergent West African
cluster
Spondweni_virus



- Download the alignment ([NEXUS format](#), [FASTA format](#))
- Phylogenetic Tree (export as [PDF](#), [NEXUS Format](#)):
- View the [PAUP* Log file](#)

Developed by: **FIOCRUZ/Bahia, Brazil** (Maria Inés Restovic, Marta Giovanetti, Vagner Fonseca, Munilo Freire, Luiz Alcantara), **KU Leuven, Belgium** (Kristof Theys, Pieter Libin, Lize Cuyper, Ana Abecasis, Anne-Mieke Vandamme), **Oxford, U.K.** (Nuno Faria and Oliver Pybus), **Evandro Chagas Institute, Brazil** (Marcio Roberto Teixeira Nunes), **CDC/OID/NCEZID** (Gilberto A. Santiago), **Emweb bvba, Belgium** (Koen Deforche) and **Africa Centre/UKZN, South Africa** (Tulio de Oliveira).

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YELLOW FEVER VIRUS TYPING TOOL

YELLOW FEVER VIRUS PHYLOGENETIC TYPING TOOL

Version 2.61

Kwazulu-Natal Research and Innovation Sequencing Platform

YELLOW FEVER VIRUS PHYLOGENETIC TYPING TOOL

This tool is designed to use Blast and phylogenetic methods in order to identify the Yellow Fever Virus genotypes of a nucleotide sequence.

Note for batch analysis: The tool accepts up to 2000 sequences at a time.

INPUT

Submit one or more sequences that are typed individually.
[Click here](#) to load some sample data.

Sequence

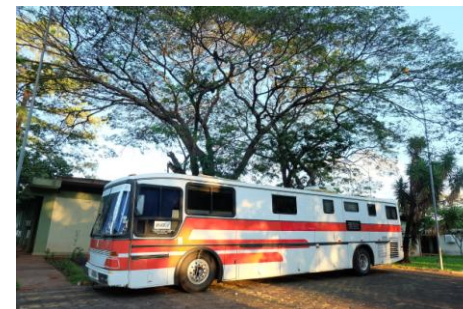
CLICK OR DROP FILE

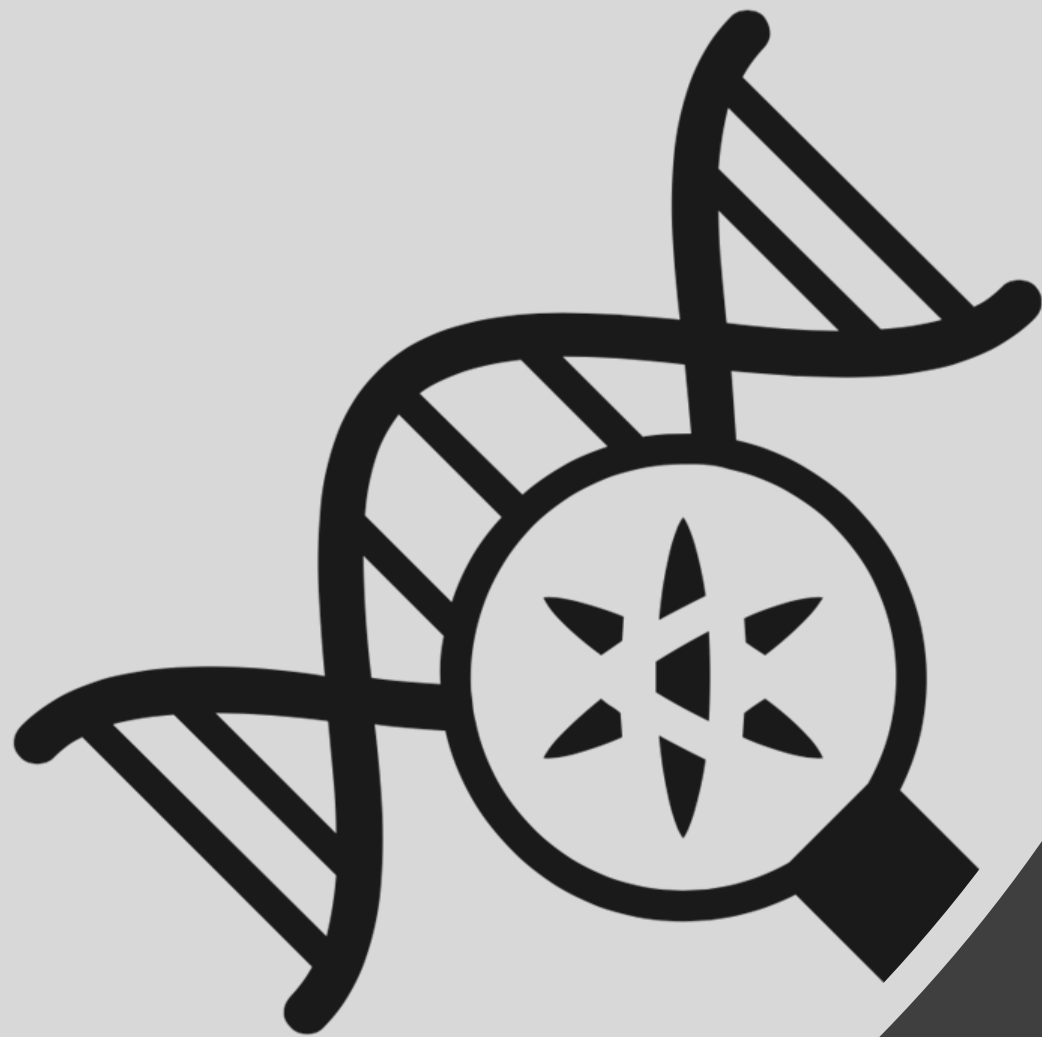
(Paste or upload a FASTA sequence)

START

CLEAR

Portable DNA sequencing in Midwest Brazil: impacting the response to the arboviral diseases on the ground, providing a snapshot of their complex dynamic's evolution.





Metagenomica

Species	NT / AA Identity (%)		Genome
Chikungunya virus	96.893	98.3888	
Chikungunya virus	96.8938	98.3888	
Chikungunya virus	96.879	98.362	
Chikungunya virus	96.8846	98.362	

Genomas completos de CHIKV de metagenomica

119 Genomas completos de CHIKV, ZIKV, DENV

Transfer technology



Public
Engagement

Education

Open Access

Gender

Ethics



Entomology



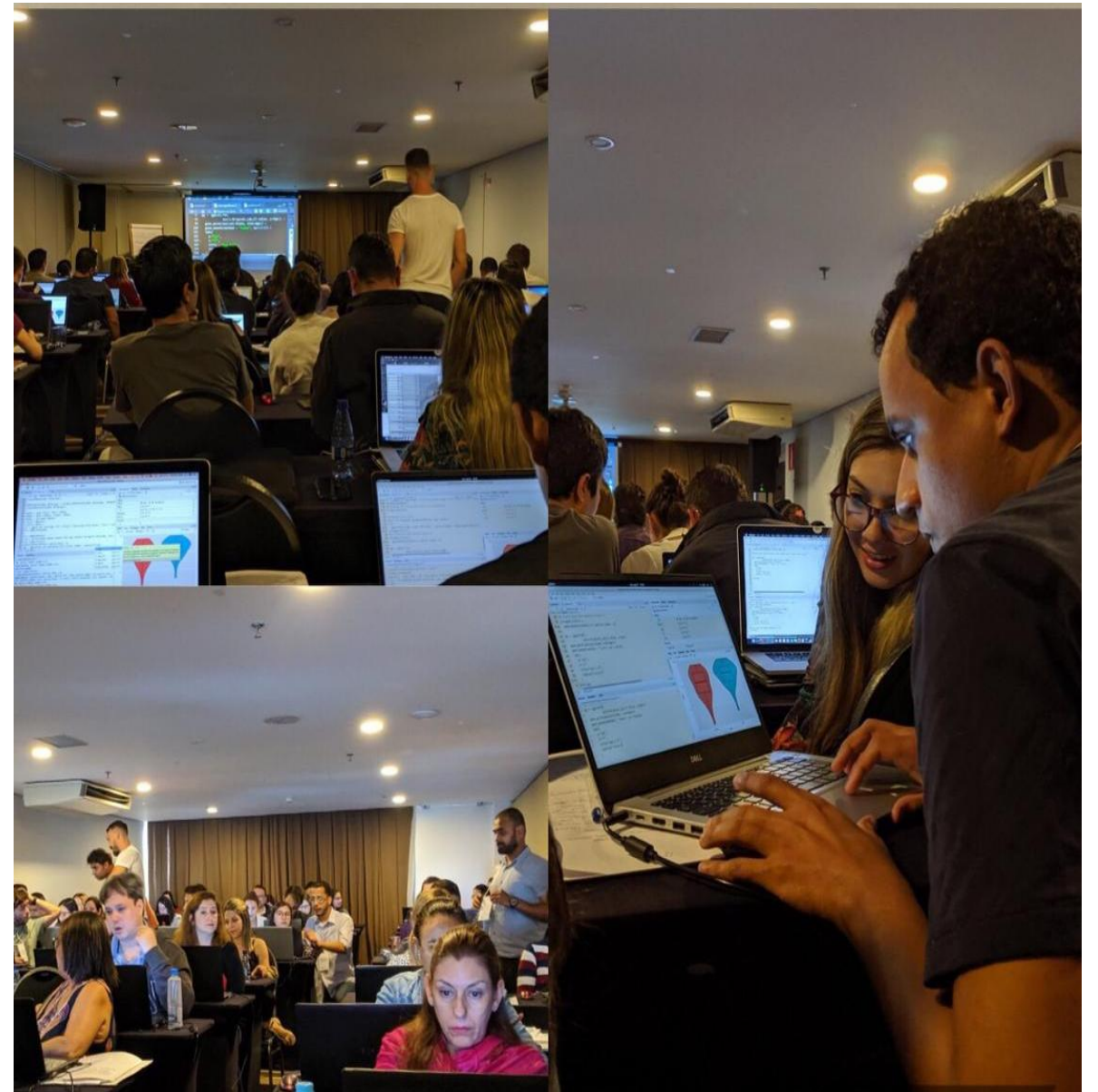
Sequencing



Bioinformatics



Coordination and logistics



What next ?

Towards a real-time genomic-informed surveillance of viral pathogens

Syndromic surveillance



Mapping disease emergence factors



- Epidemiological data
- Pathogen samples:
 - Patients
 - Sentinel population
 - Domestic animals
 - Wildlife
 - Environment



LACEN (2016)

Outbreak isolate

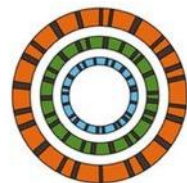
Control isolate

DNA sequencing



De novo assembly of sequences

Mapping to reference genome

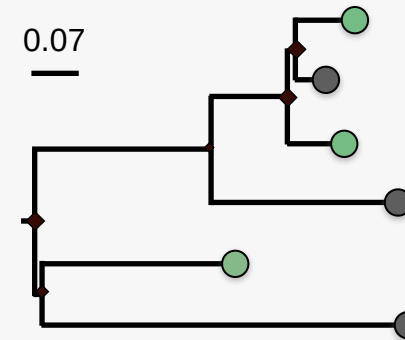


AGACTAATTGCGCT
AGACTAATTGCGCT
AGACTAATTGCGCT
AGACTAATTGCGCT
AGACTAATTGCGCT

Identification of high quality variants

Comparative analysis and visualization of relationships

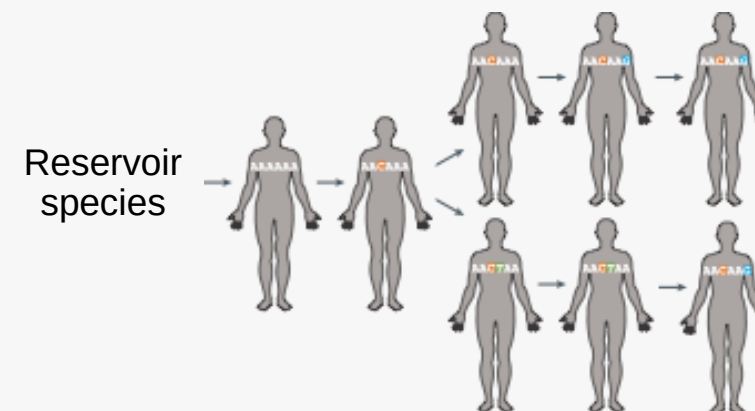
0.07



- Genome from human
- Genome from reservoir/vector
- ◆ Clade and trait (geography & host) statistical probabilities

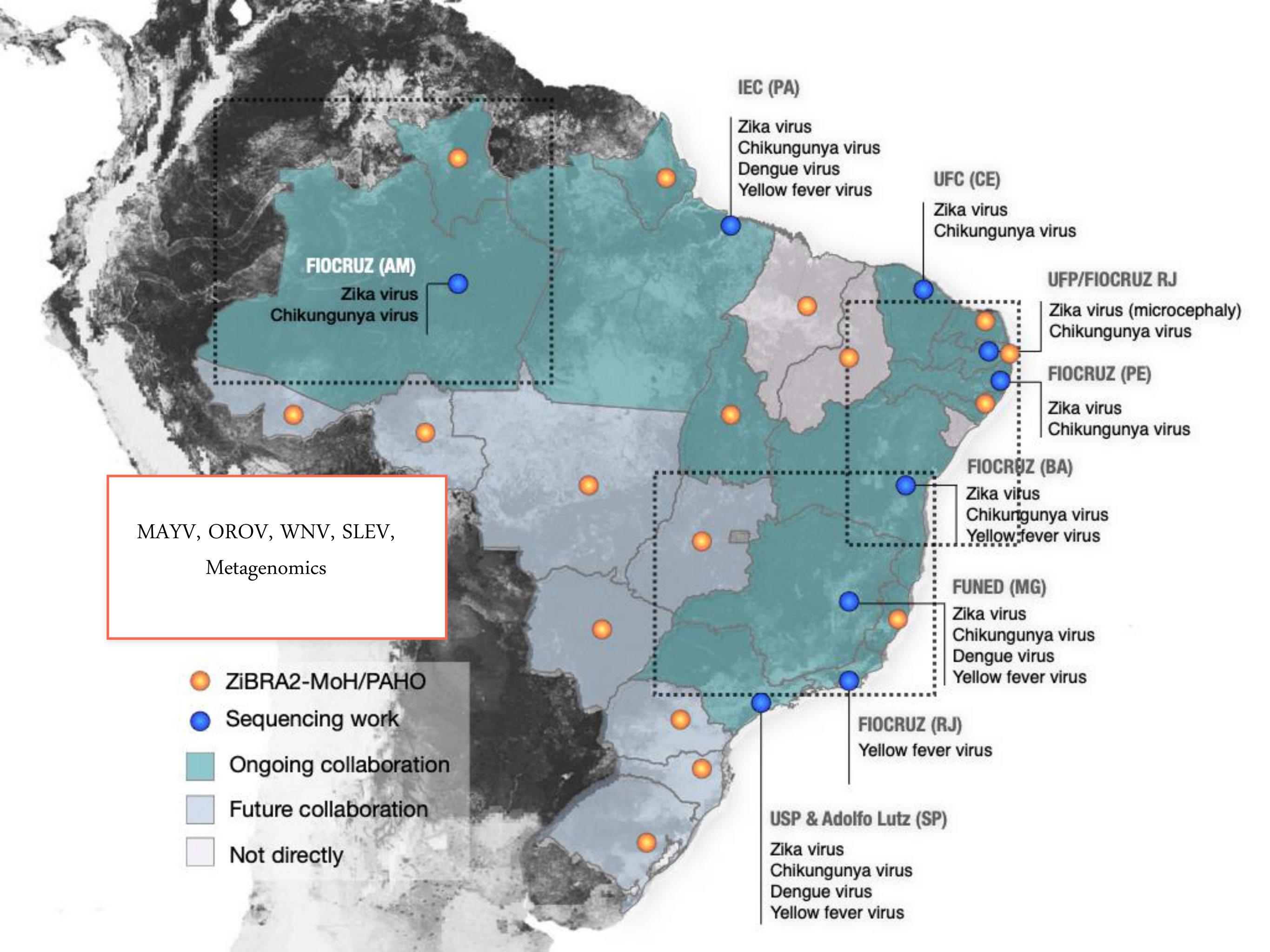
Phylogenetic inference

Epidemiological reconstruction



Transmission inference

Adapted from
Gardy and Loman, *Nature Rev Genet* (2018)



Mobile Real-time genomic of arboviruses in Brazil

Marta Giovanetti

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ALAG – MENDONZA (AR) 2019

