



UZ  
LEUVEN

KU LEUVEN



# Information session of the National Reference Center for Rotavirus

**14-12-2023**

Jelle Matthijnsens

Lize Cuypers

# Recap important information

- **Accreditation** requested: please enter your name (and RIZIV/INAMI if applicable) in the chat box
- **Interactive sessions:** you can speak up by unmuting your microphone to ask questions or raise comments in the chat box
- No recording of the session but slides will be shared  
<https://www.uzleuven.be/nl/laboratoriumgeneeskunde/nationale-referentiecentra-en-referentielaboratoria>  
<https://www.sciensano.be/nl/nrc-nrl/nationaal-referentiecentrum-nrc-voor-rotavirus>



The screenshot shows a website page with the UZ Leuven logo in the top right corner. A blue navigation bar contains the text 'Laboratoriumgeneeskunde'. Below the navigation bar, the breadcrumb trail reads 'Home > Diensten, centra en afdelingen > Laboratoriumgeneeskunde'. The main heading is 'Informatiesessies nationale referentiecentra voor humane microbiologie'. A list of sessions is provided below:

- Streptococcus pneumoniae (invasief) - 30 maart 2023
- Enterovirussen (inclusief poliovirus en parechovirus) en rioolwaterscreening - 27 april 2023
- Mycosis (UZ Leuven en CHU Liège) - 8 Juni
- NRC respiratory pathogens (UZ Leuven en UZA) - 26 oktober

## Rotavirus

- RIZIV nomenclature and/or invoice
  - Rotavirus antigen test (EIA): Rotaclone®
  - Part of gastro-intestinal panel: detection of 24 pathogens (years 2022 and 2023: PR of 3.1% for rotavirus, with >50% strongly positive)
- NRC budget
  - Typing: RT-PCR and sequencing

Rotavirus  
Surveillance  
Network  
Belgium



### Bacteriën:

Campylobacter spp. (Campylobacter jejuni, Campylobacter upsaliensis, Campylobacter coli), Salmonella spp., Clostridium difficile (tcdA/tcdB), Yersinia enterocolitica, Enterotoxinogene E. coli (ETEC), Enteropathogene E. coli (EPEC), Enteroaggregatieve E. coli (EAEC), Shiga-like toxine producerende E. coli (STEC) serotype O157:H7//stx1/stx2, Entero-invasieve E. coli (EIEC)/Shigella, Plesiomonas shigelloides, Vibrio vulnificus, Vibrio parahaemolyticus, Vibrio cholerae

### Parasieten:

Entamoeba histolytica, Cryptosporidium spp., Giardia lamblia, Cyclospora cayetanensis,

### Virussen:

Humaan adenovirus F40/F41, Norovirus GI, Norovirus GII, **Rotavirus A**, Astrovirus en Sapovirus (GI, GII, GIV en GV).

## Nationaal Referentiecentrum (NRC) voor Rotavirus

### Nuttige links:

Onderaan deze pagina kan u de **NRC-rapporten** terugvinden.

De bijhorende **epidemiologische surveillance-rapporten** kan u consulteren via:

[Gezondheidsonderwerp Rotavirus](#)

[Gezondheidsonderwerp Vaccineerbare ziekte](#)

### Beschikbare testen

1. [Rotavirus Antigeen-ELISA \(RIZIV nomenclatuur\)](#)
2. [Rotavirus RT-PCR en sequentiebepaling \(referentiecentrum activiteit\)](#)

### Verantwoordelijke laboratoria

#### Coördinator

- [UZ Leuven/KU Leuven](#)



### Erkend door

- [National Institute for Health and Disability Insurance \(INAMI-RIZIV\)](#)

### Aanvraagformulieren

- [Aanvraagformulier Rotavirus](#)

Information on  
vaccination status  
and in case of  
outbreak!

Jaar	Beschrijving
2022	 <a href="#">Rotavirus Report 2021-2022 (English only)</a>
2021	 <a href="#">Rotavirus report 2020-2021</a>

# NRC Rotavirus

Jelle Matthijnsens

Marc Van Ranst

Elke Wollants

Mandy Bloemen

Lize Cuypers

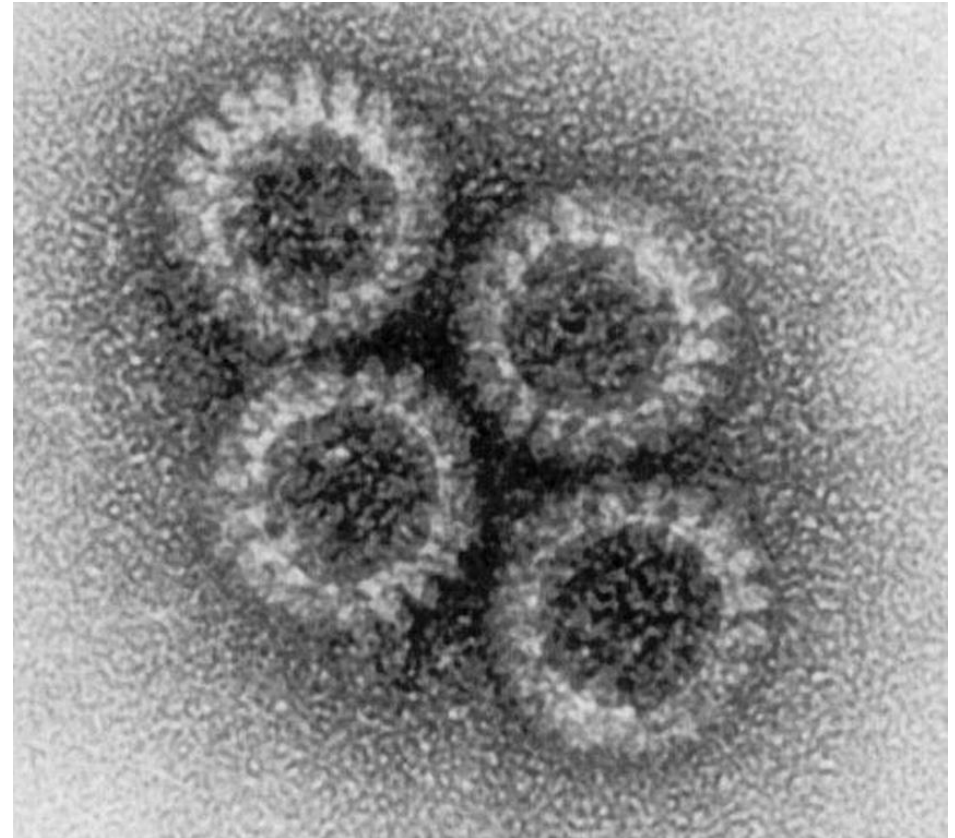
# Overview

- Introduction
- Rotavirus surveillance UZ Leuven (1981-2023)
- Rotavirus surveillance NRC (2009-2023)
- Rotavirus co-infections
- Vaccine-derived rotavirus strains

# Introduction

# Rotavirus Characteristics

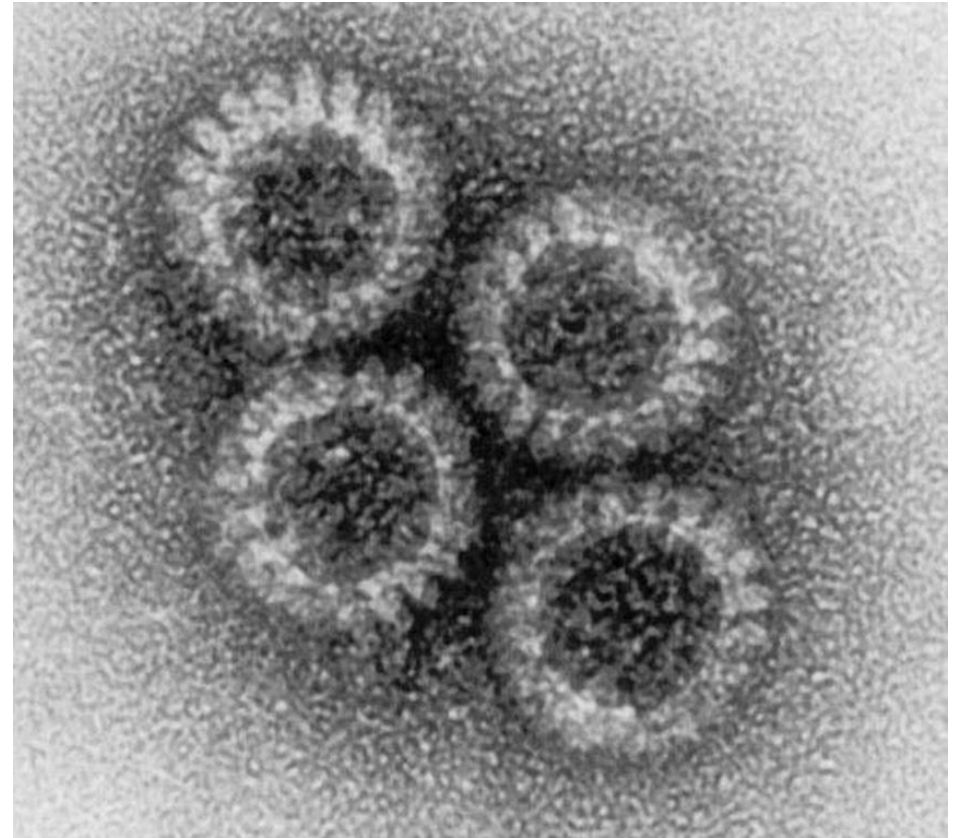
- 'Rotavirus' derived from 'rota' meaning 'wheel' in latin





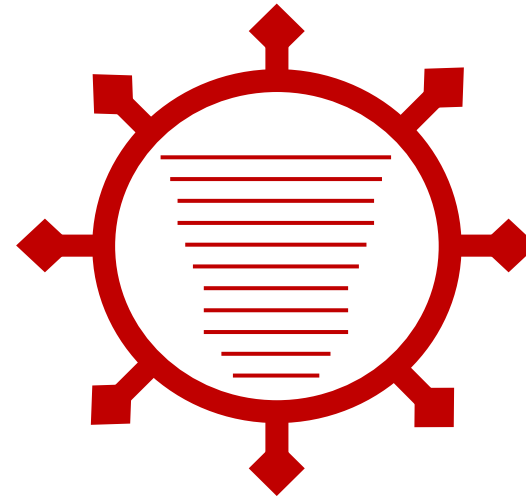
# Rotavirus Characteristics

- 'Rotavirus' derived from 'rota' meaning 'wheel' in latin
- **No envelope**



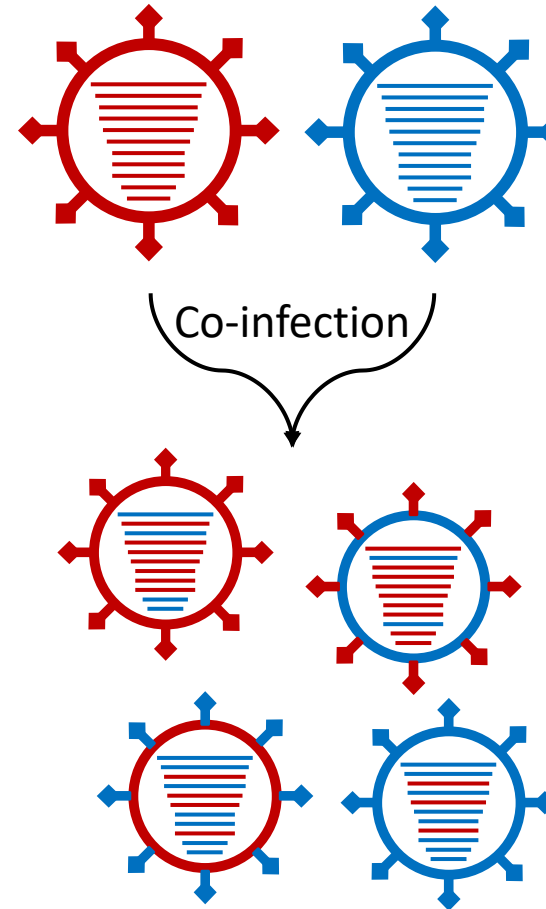
# Rotavirus Characteristics

- 'Rotavirus' derived from 'rota' meaning 'wheel' in latin
- No envelope
- **Genome comprised of 11 segments of dsRNA**



# Rotavirus Characteristics

- 'Rotavirus' derived from 'rota' meaning 'wheel' in latin
- No envelope
- Genome comprised of 11 segments of dsRNA
- **Reassortment**



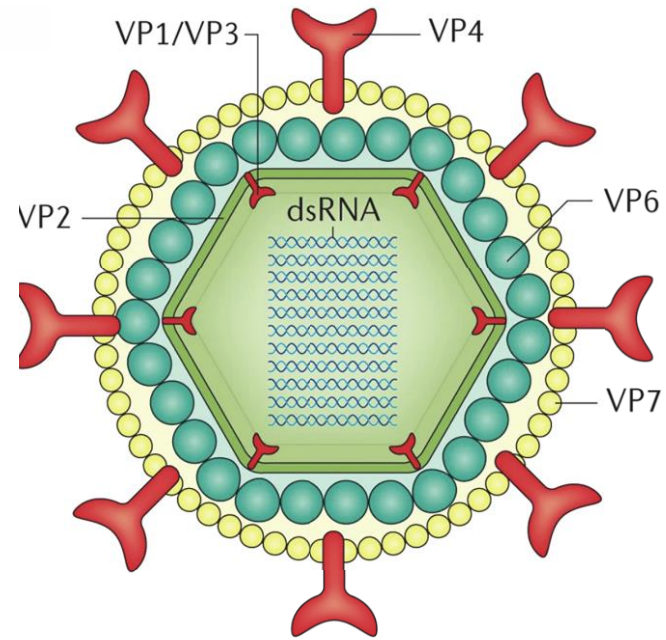
# Rotavirus Characteristics

- 'Rotavirus' derived from 'rota' meaning 'wheel' in latin
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- Genome comprised of 11 segments of dsRNA
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- Family *Sedoreoviridae*
- Genus *Rotavirus*
- Species *Rotavirus A-D* and *F-J*



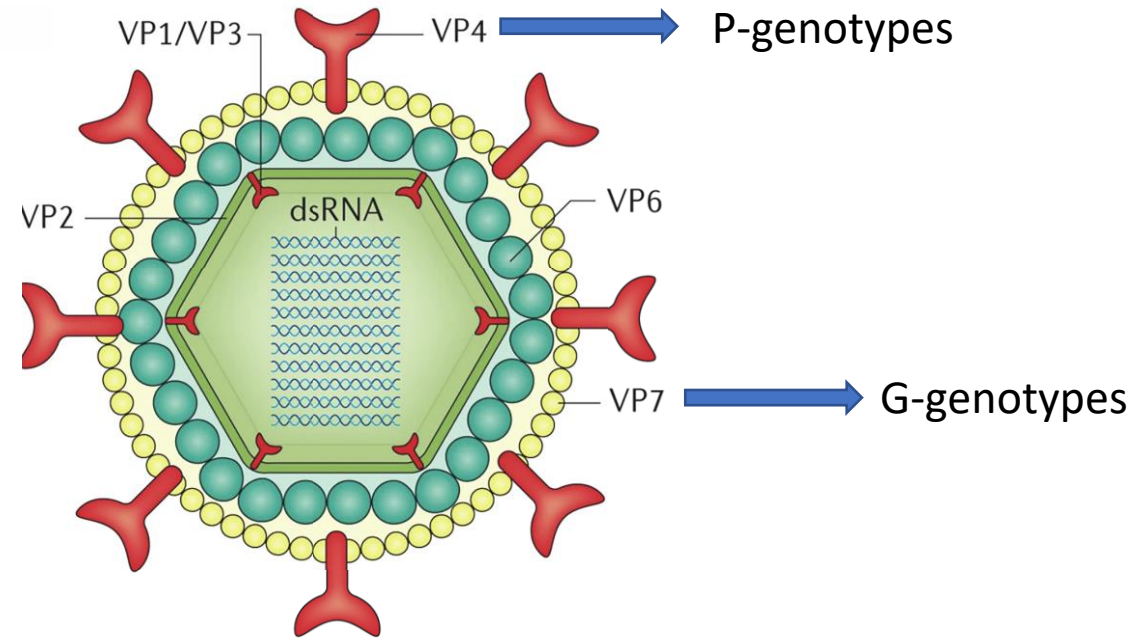
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- **Rotavirus A: G- and P-genotypes**



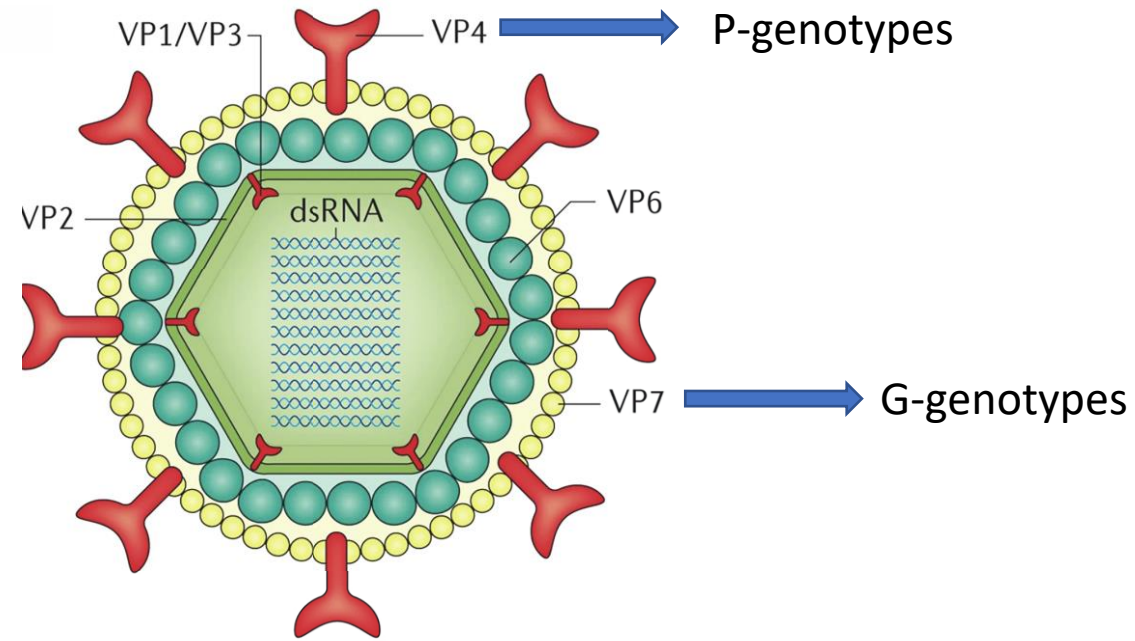
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- **Rotavirus A: G- and P-genotypes**



VP7	VP4	VP6	VP1	VP2	VP3	NSP1	NSP2	NSP3	NSP4	NSP5	
Gx	P[x]	Ix	Rx	Cx	Mx	Ax	Nx	Tx	Ex	Hx	
G1	P[8]	I1	R1	C1	M1	A1	N1	T1	E1	H1	Wa
G2	P[4]	I2	R2	C2	M2	A2	N2	T2	E2	H2	DS-1

# Rotavirus Vaccines

- Live-attenuated / Oral
- Administered @ 2, 4 (6) months
- Safe and effective

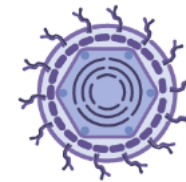


# Rotavirus Vaccines

- Live-attenuated / Oral
- Administered @ 2, 4 (6) months
- Safe and effective
- Licenced worldwide
  - Rotarix (2 doses): Monovalent



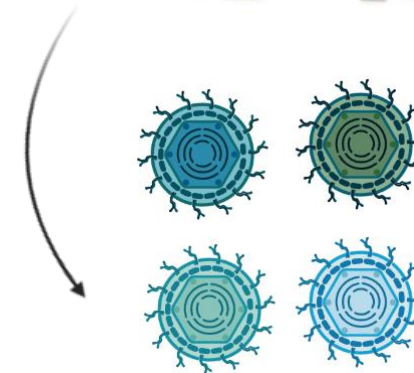
Attenuated  
Human RVA



GlaxoSmithKline (GSK)

# Rotavirus Vaccines

- Live-attenuated / Oral
- Administered @ 2, 4 (6) months
- Safe and effective
- Licenced worldwide
  - Rotarix (2 doses): Monovalent
  - RotaTeq (3 doses): Pentavalent

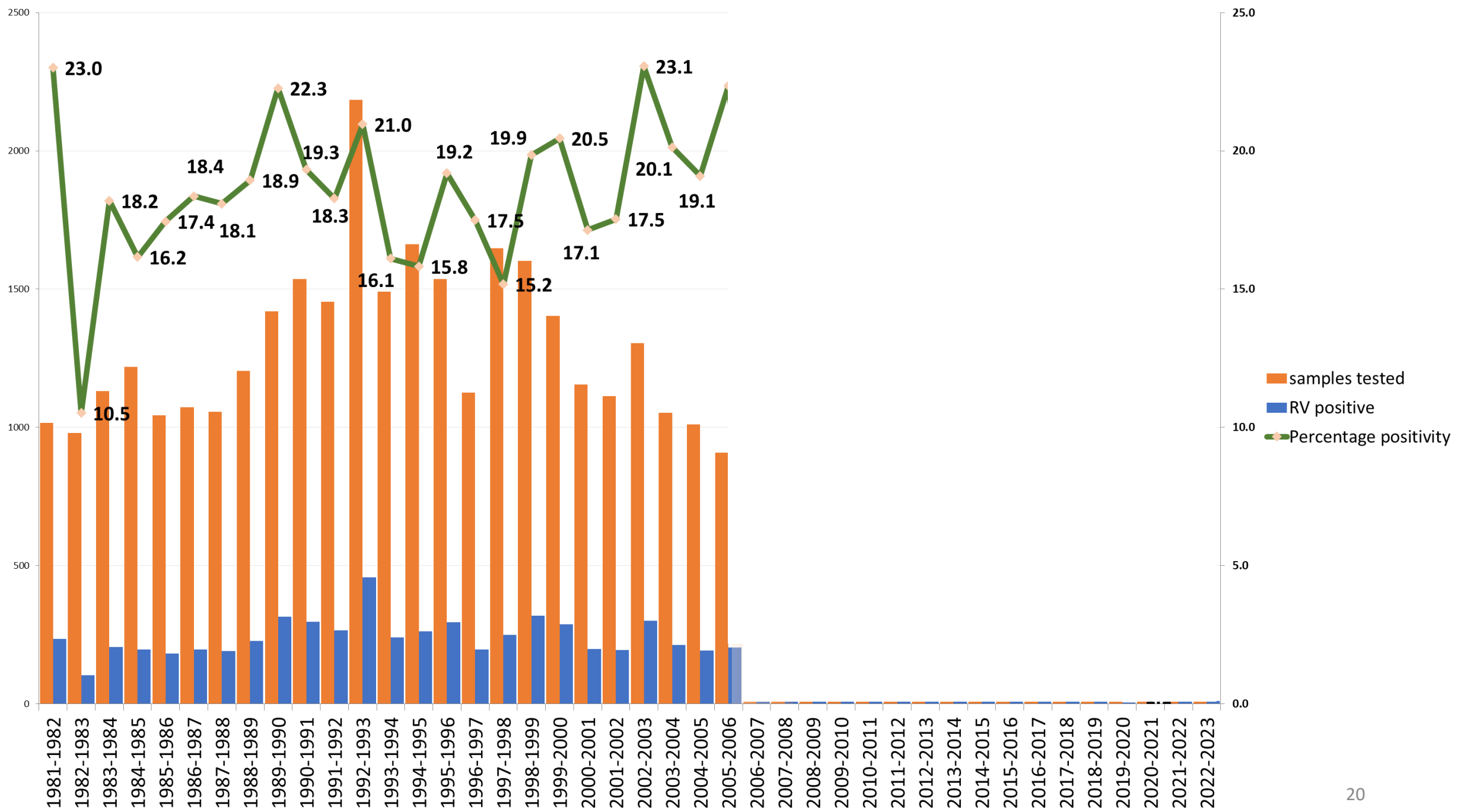


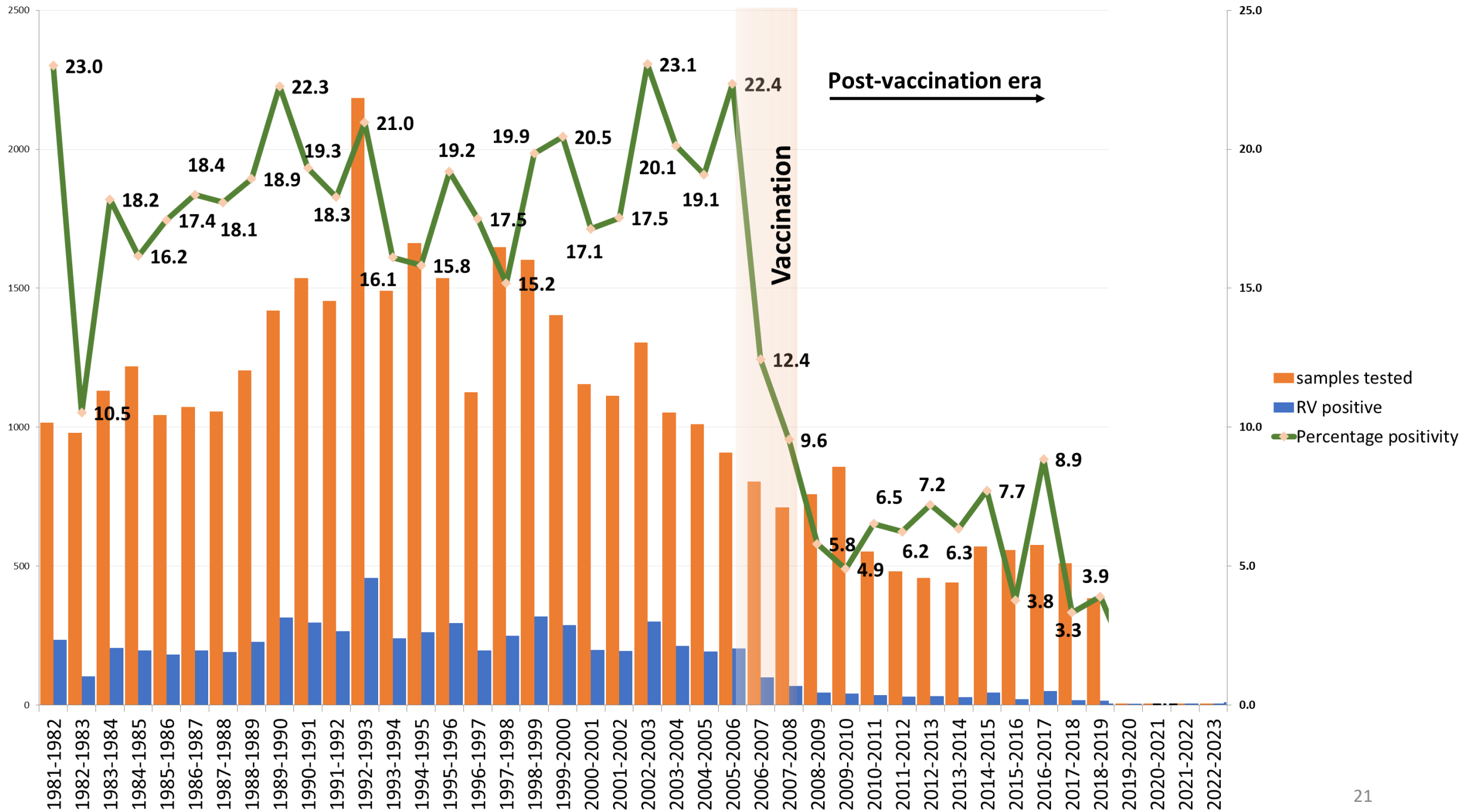
5 reassortant  
Bovine x Human RVA

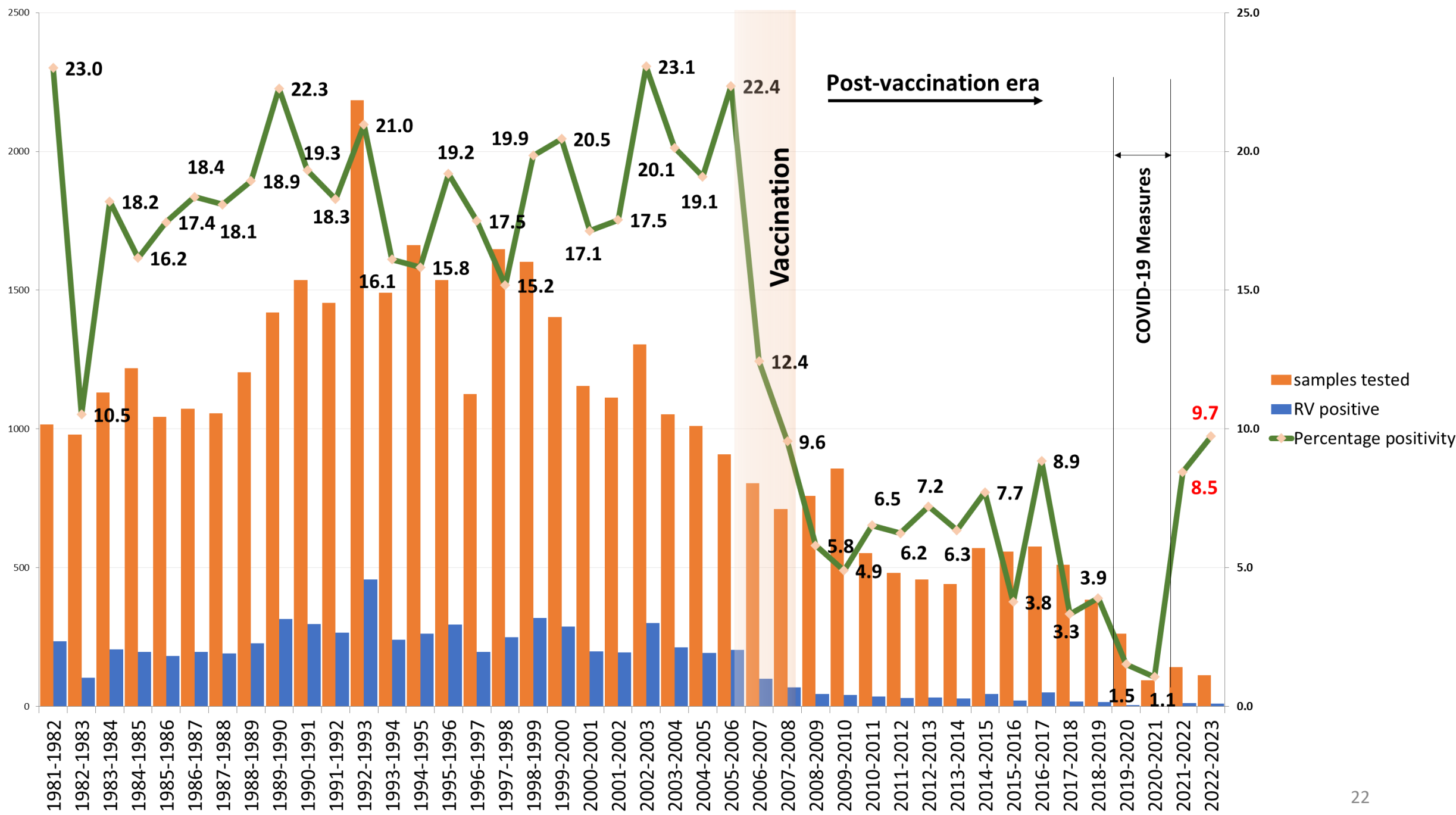


Merck

# Rotavirus surveillance UZ Leuven (1981-2023)







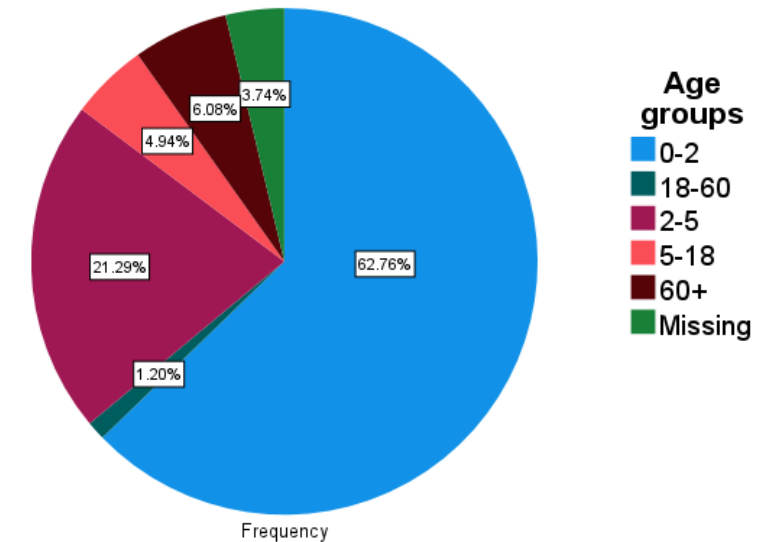
# Rotavirus surveillance NRC (2009-2023)

# NRC for Rotavirus A, 2009-2023

More than >8000 samples over the course of 14 years

Season: August 1 → July 31

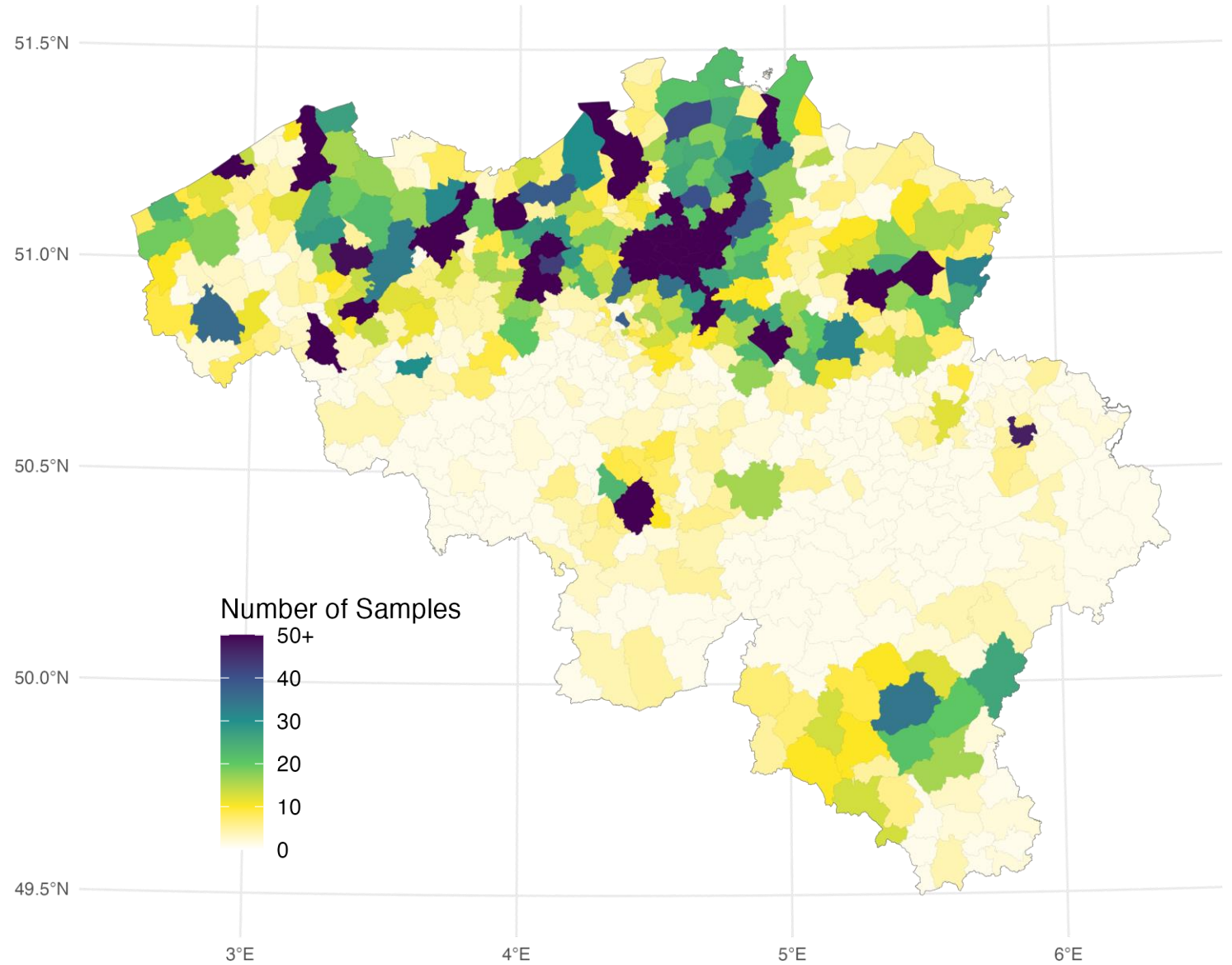
Age information (completeness)



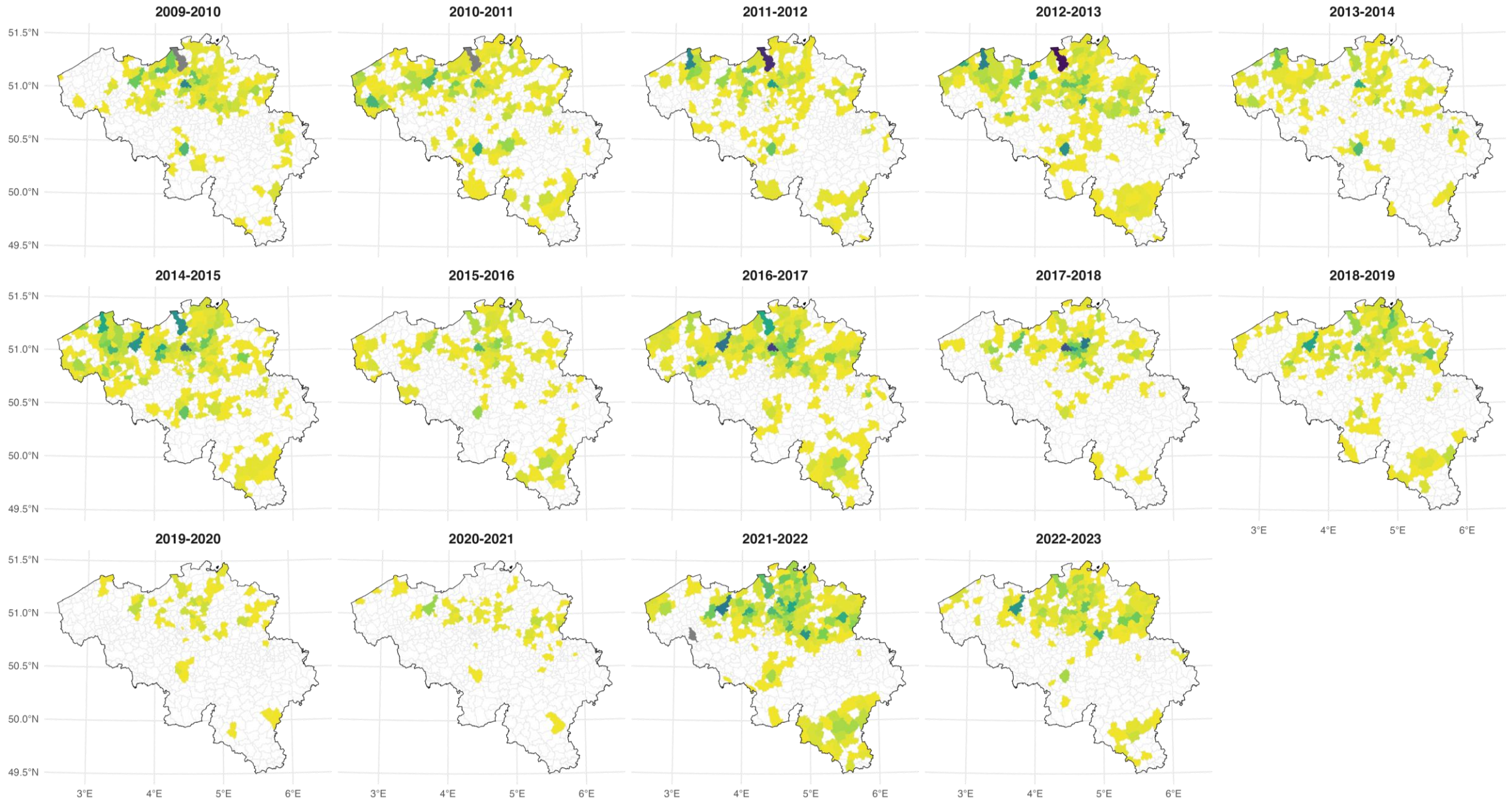


# Overall sample distribution

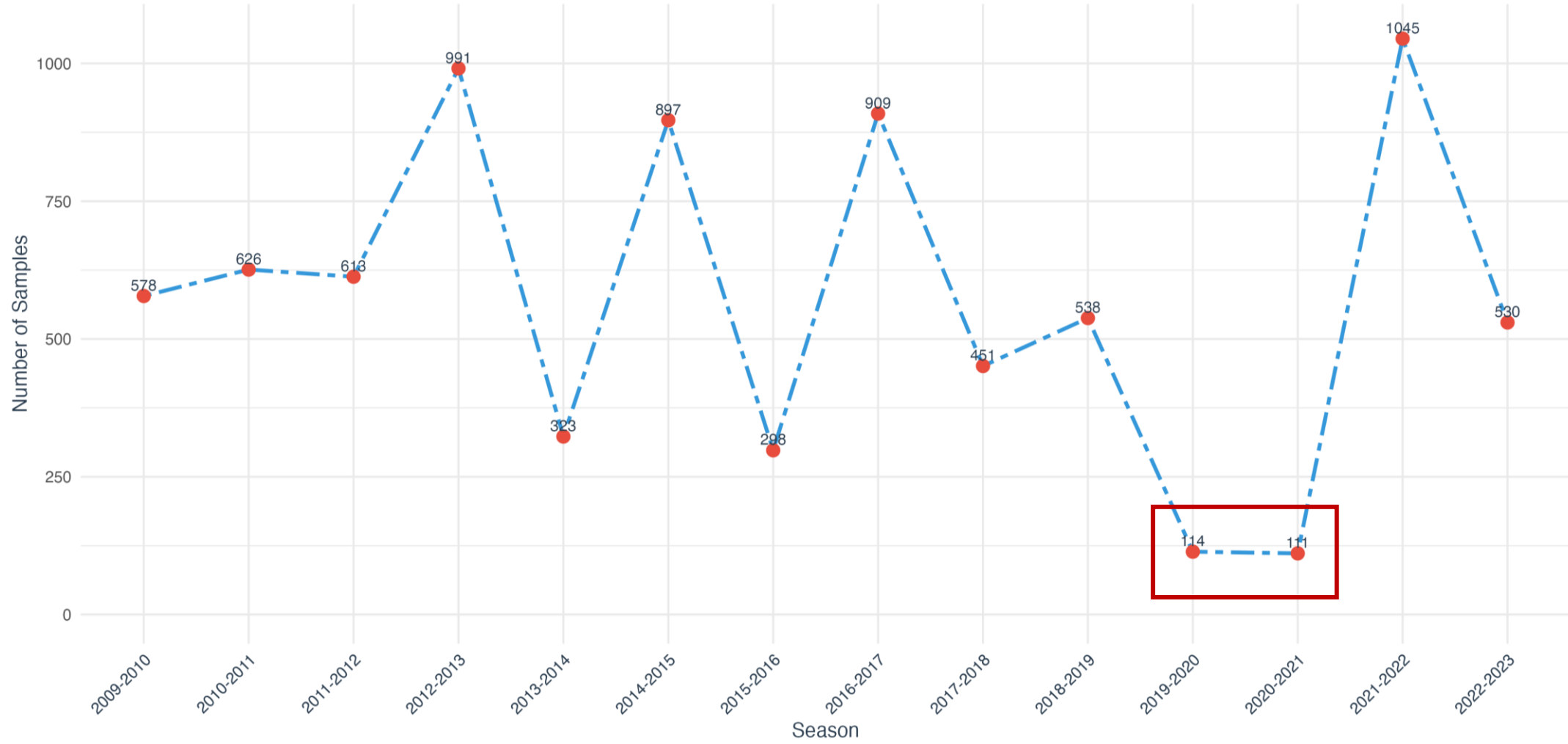
All seasons including 2009-2010 and 2022-2023



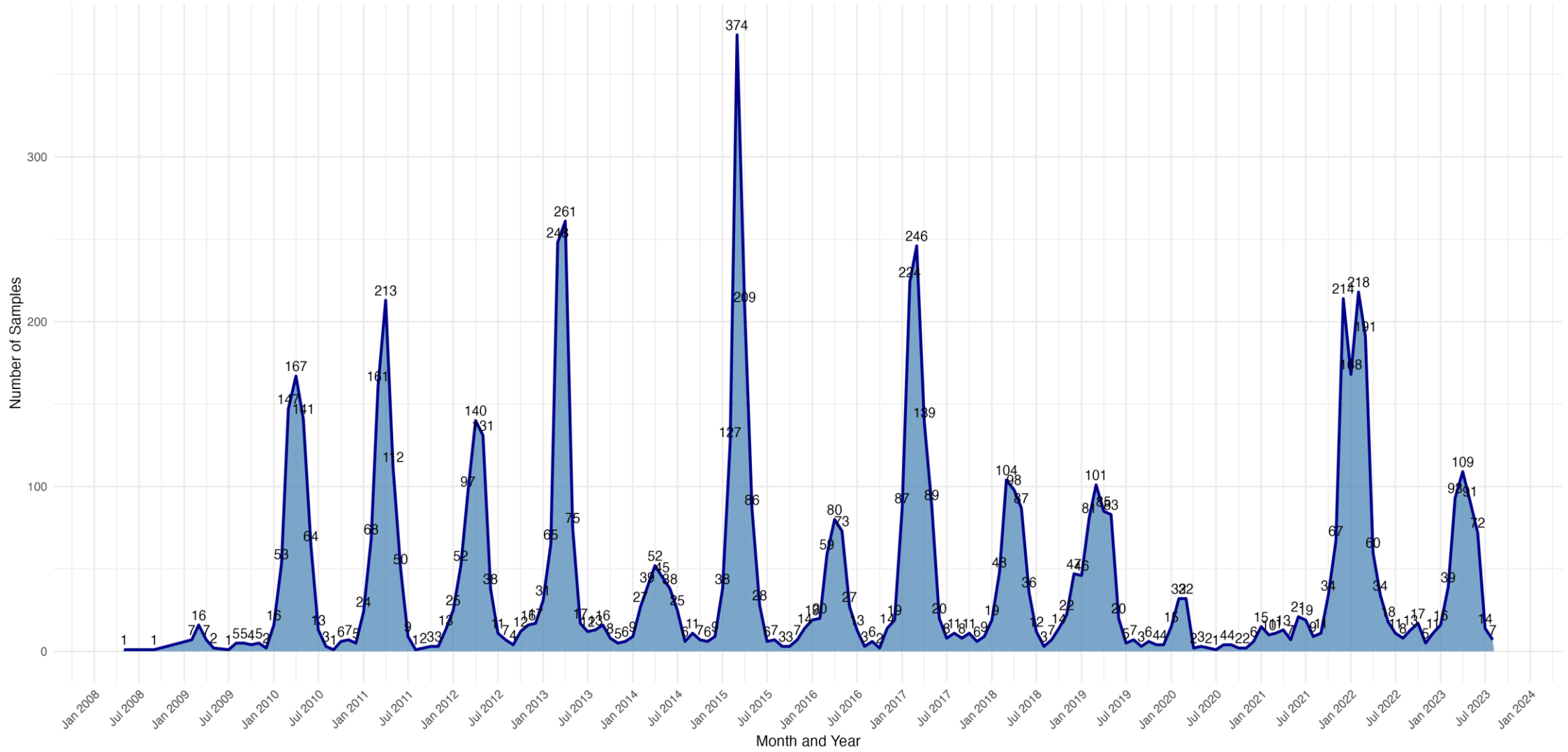
Sample Distribution in Belgium



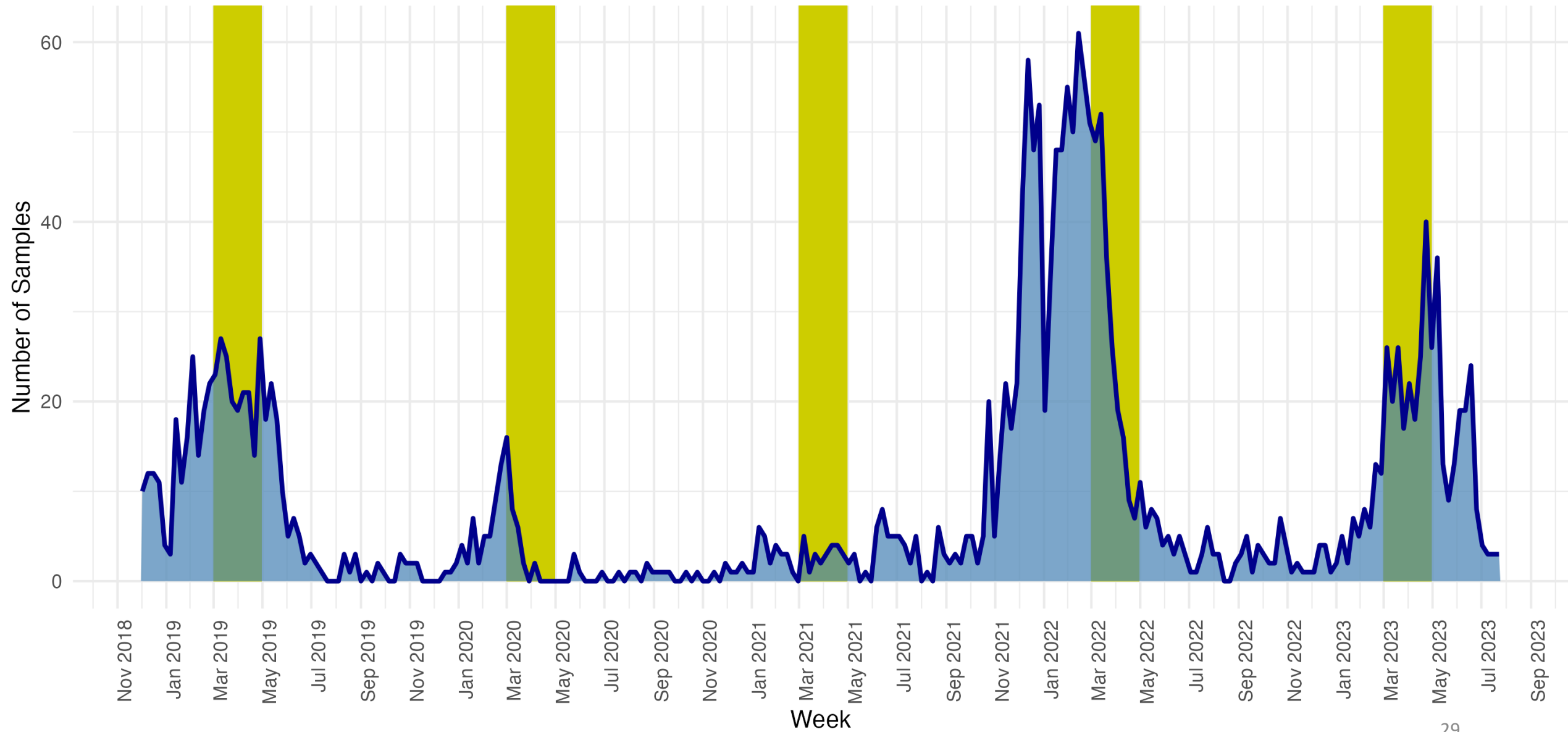
# Sample counts per year

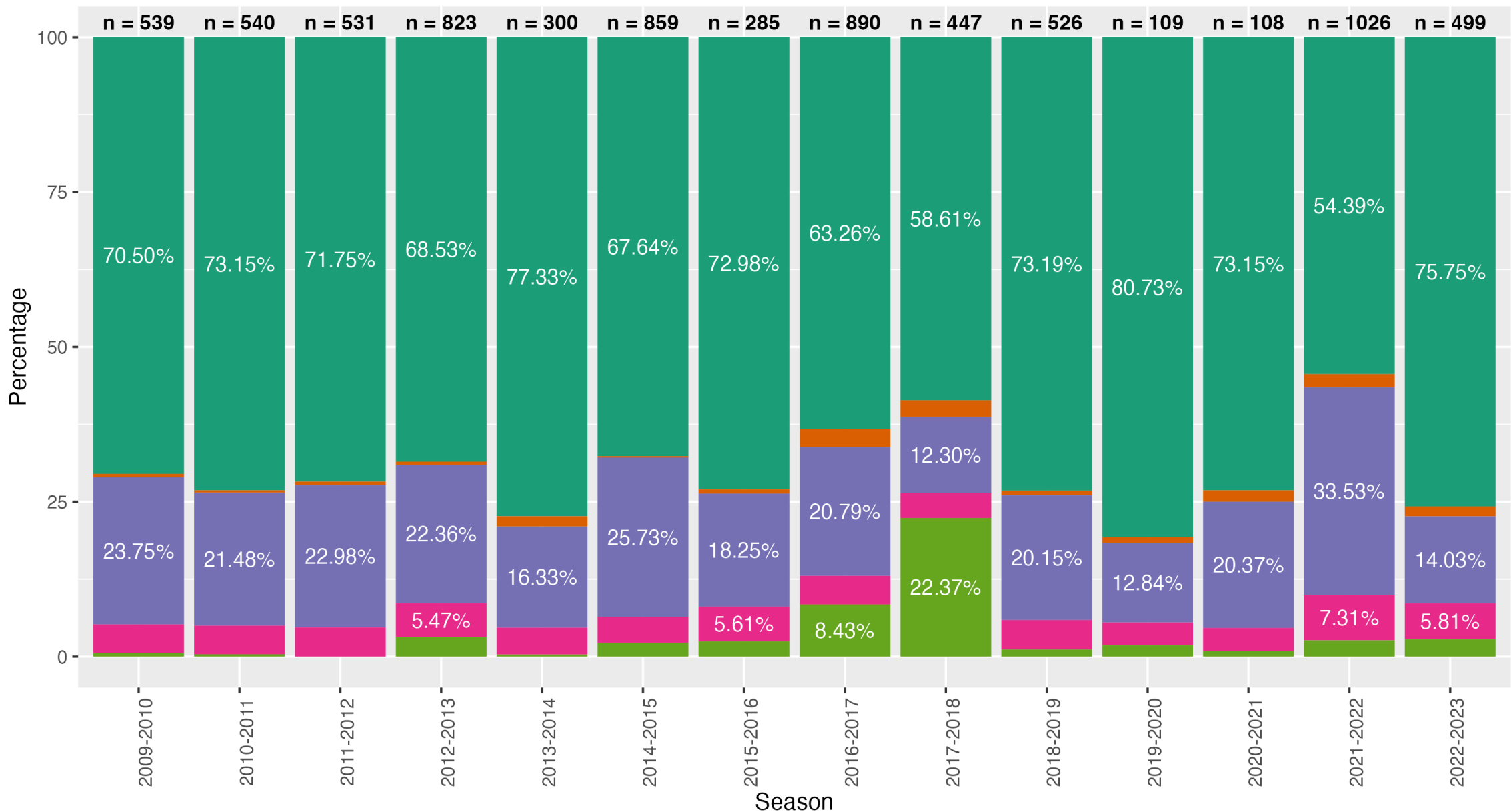


Number of Samples Collected Over Months

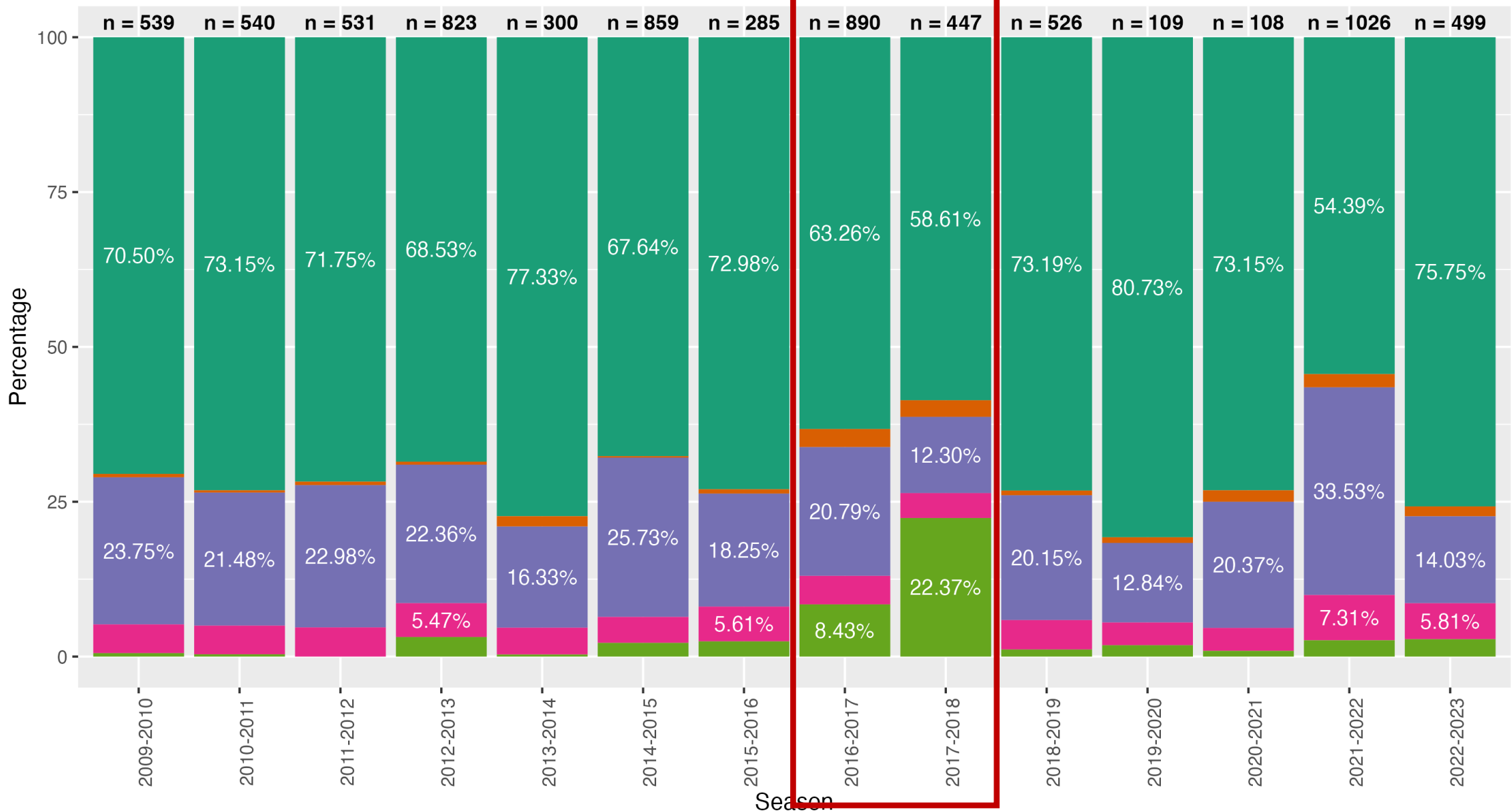


# Number of Samples Collected Over Weeks



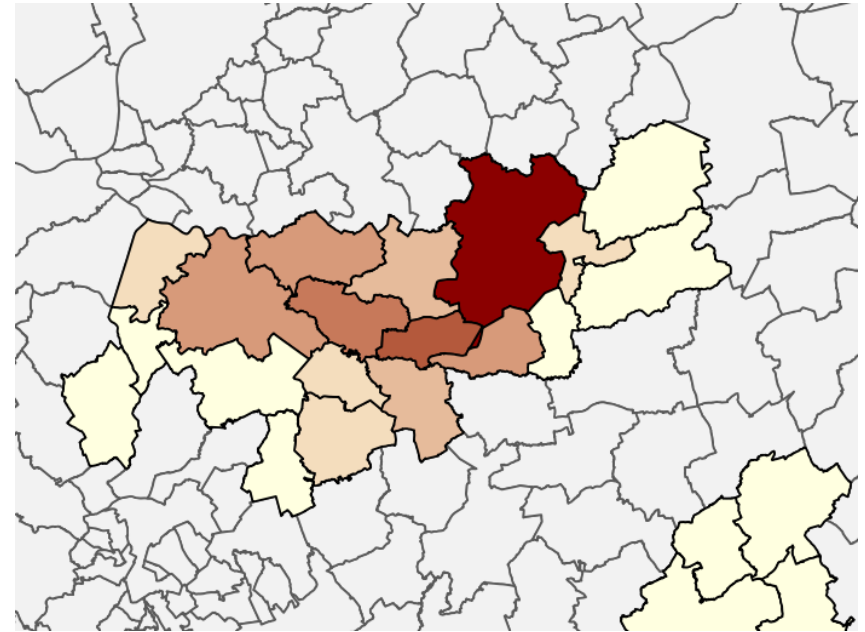
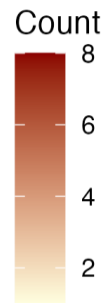
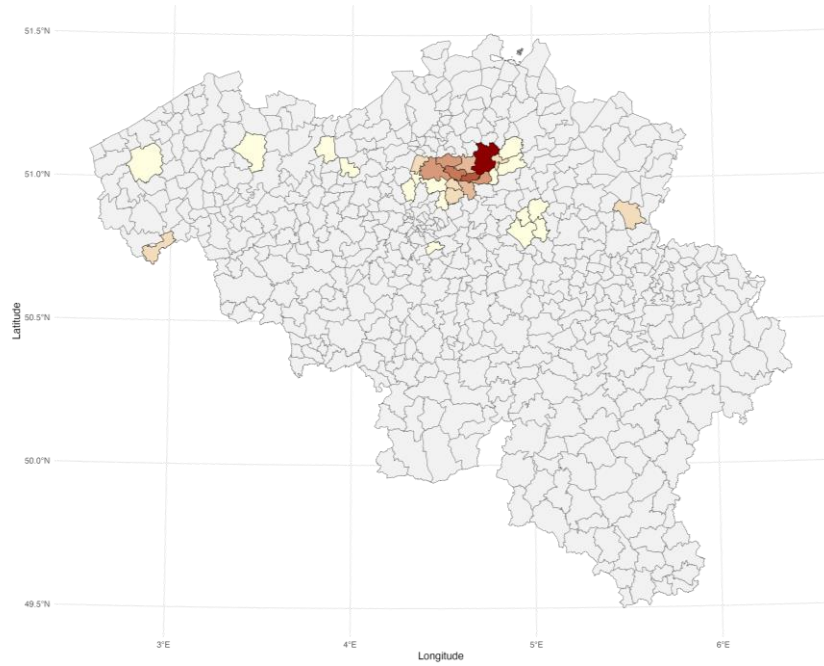


Age Group 0-2 18-60 2-5 5-18 60+

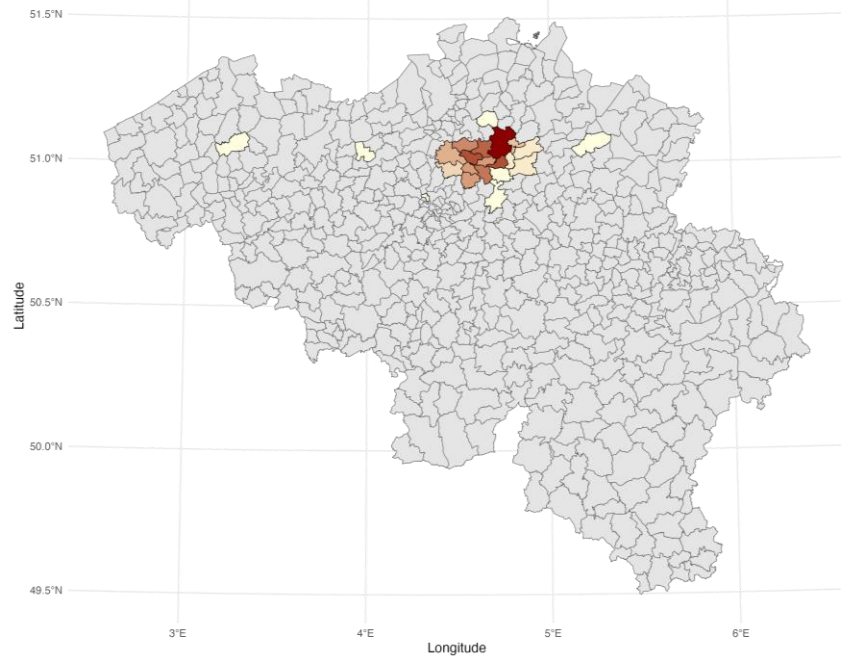


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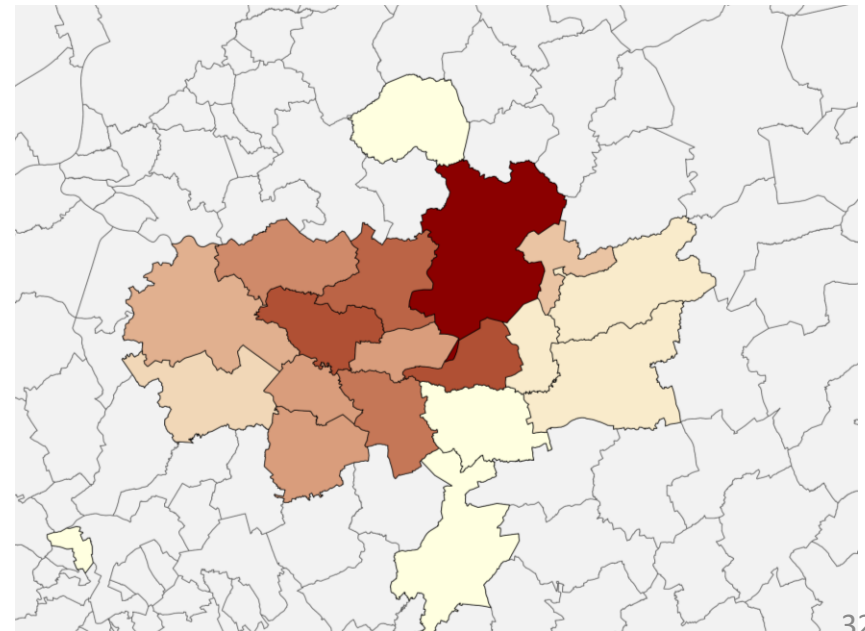
Data for 2016-2017 Season



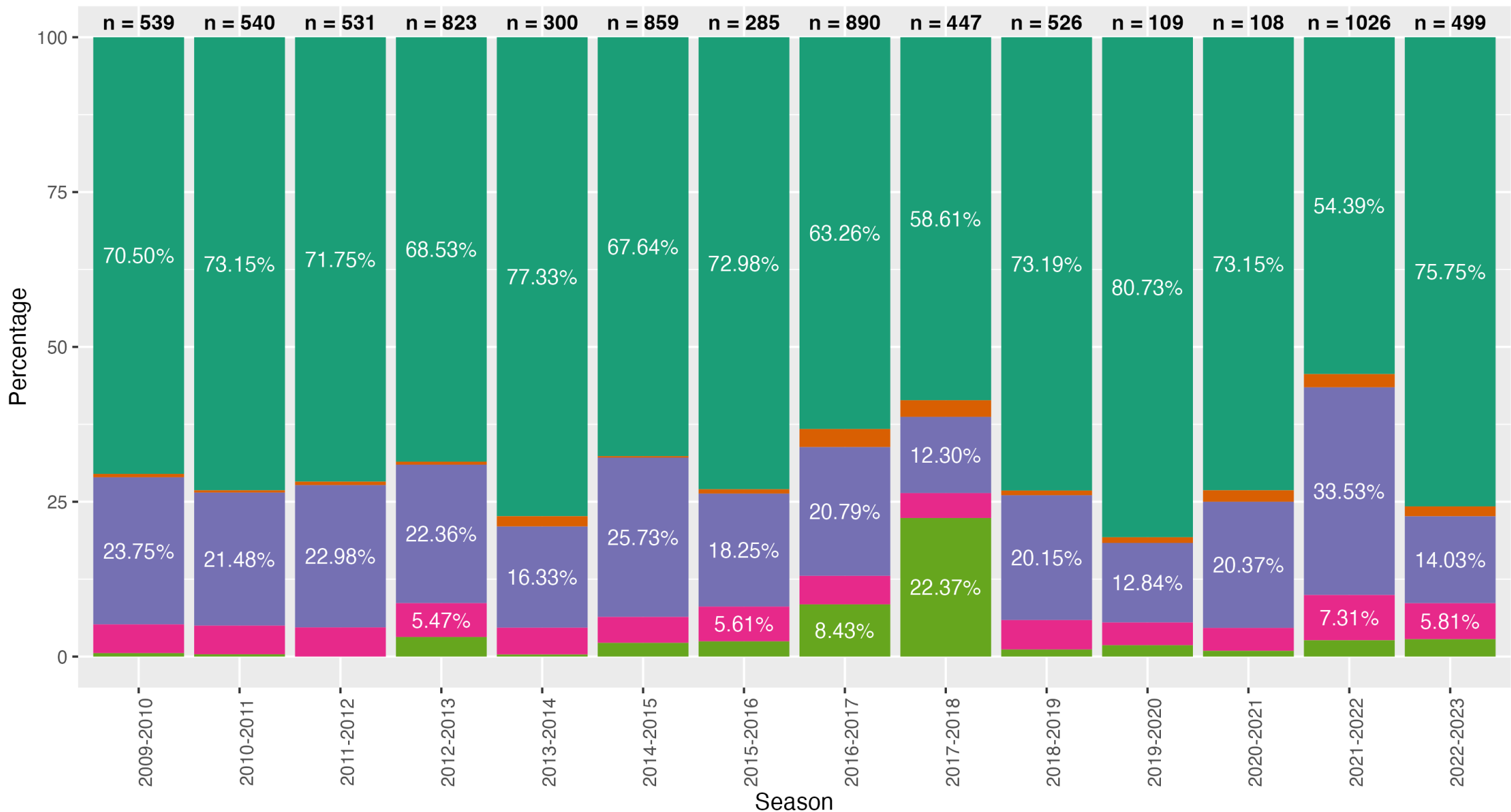
Data for 2017-2018 Season (Age Group 60+)



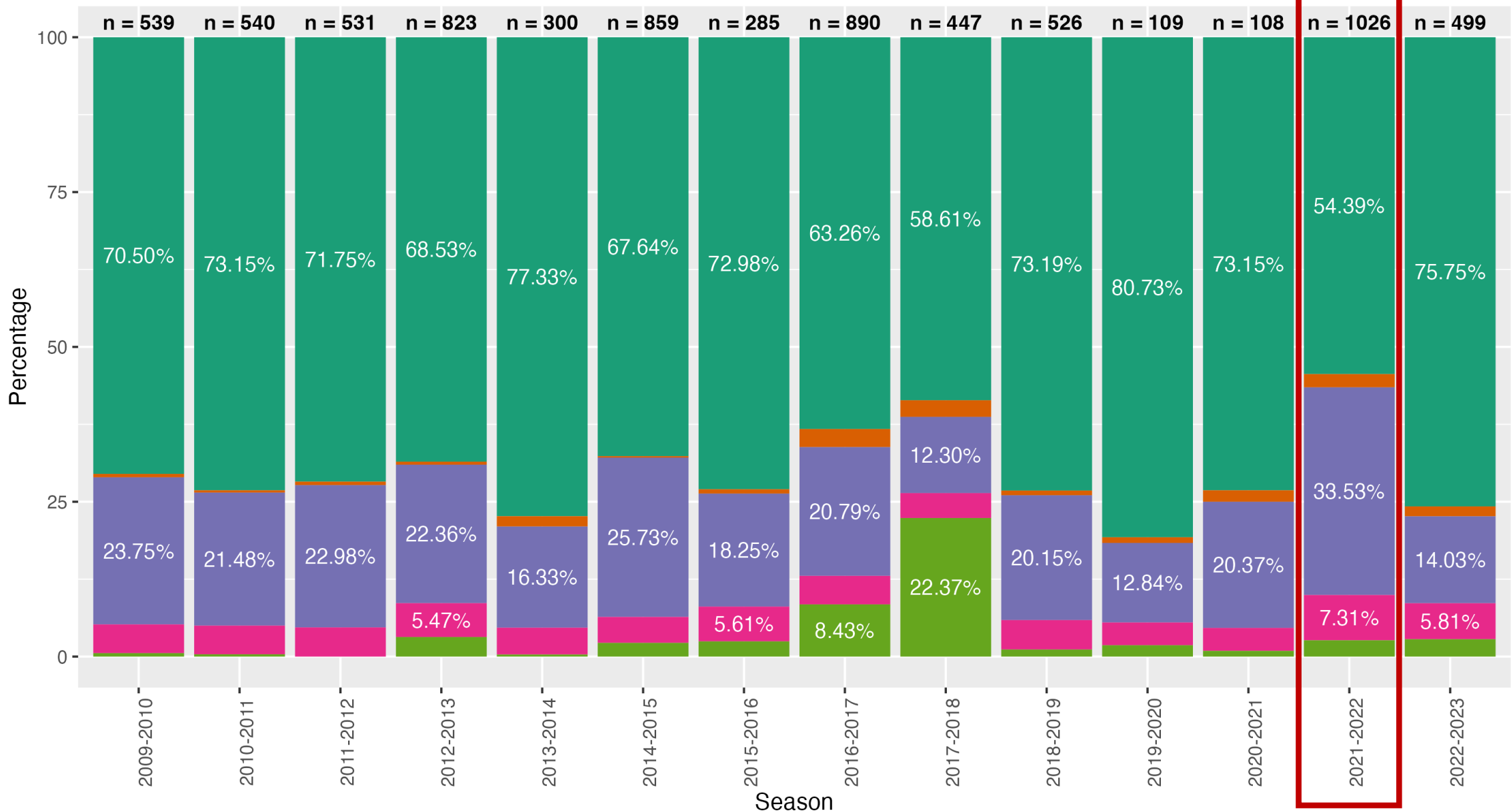
G2P[4]  
genotype



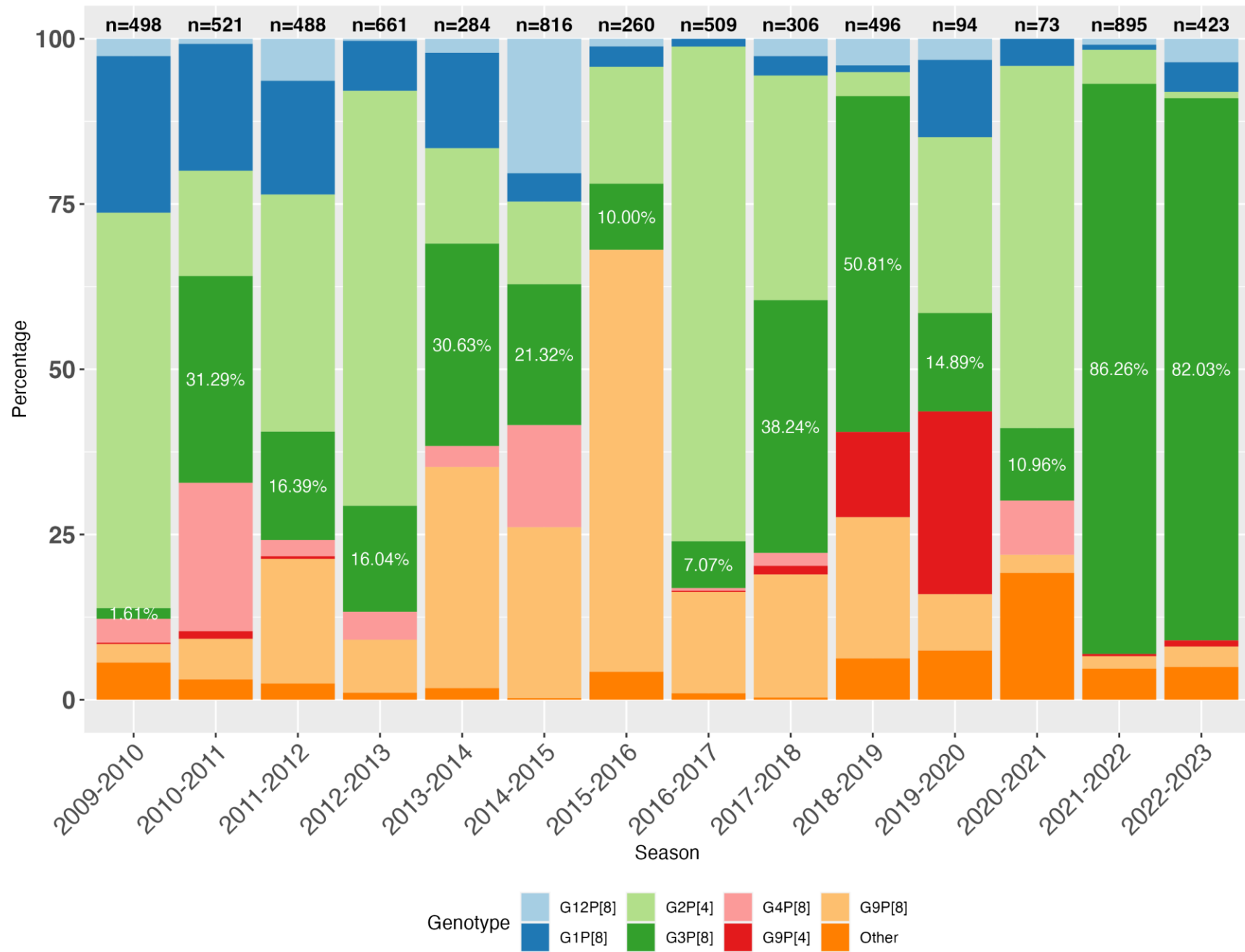


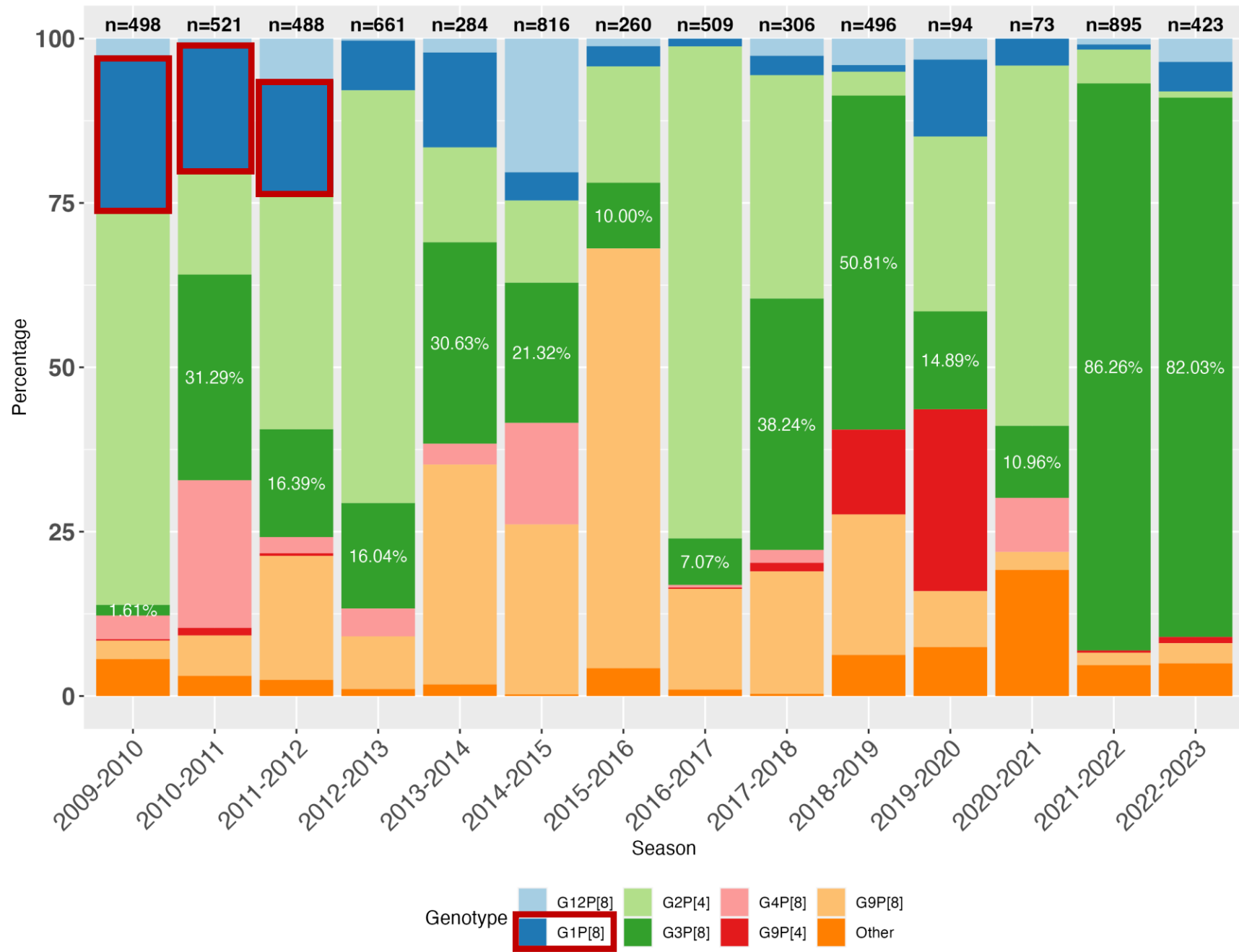


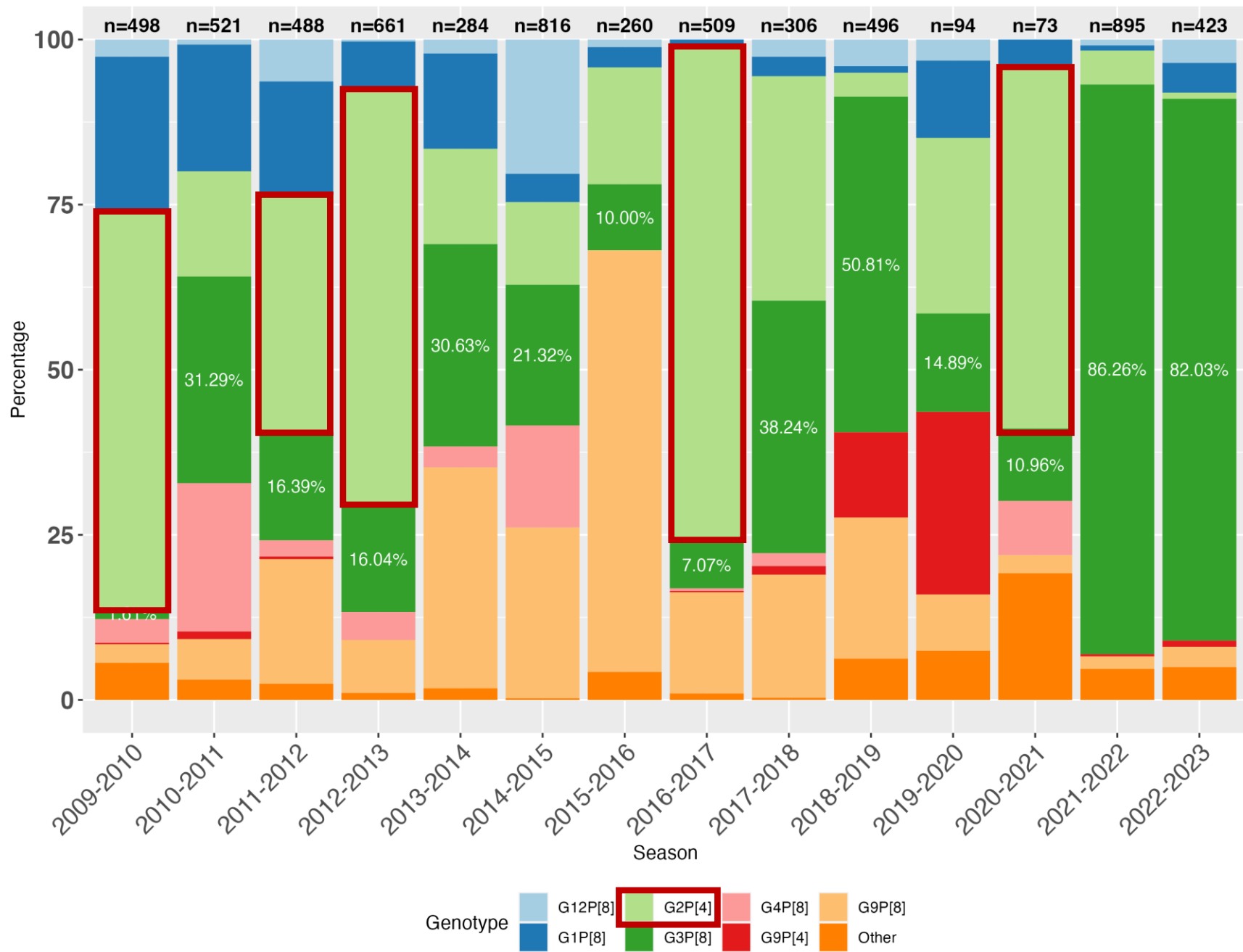
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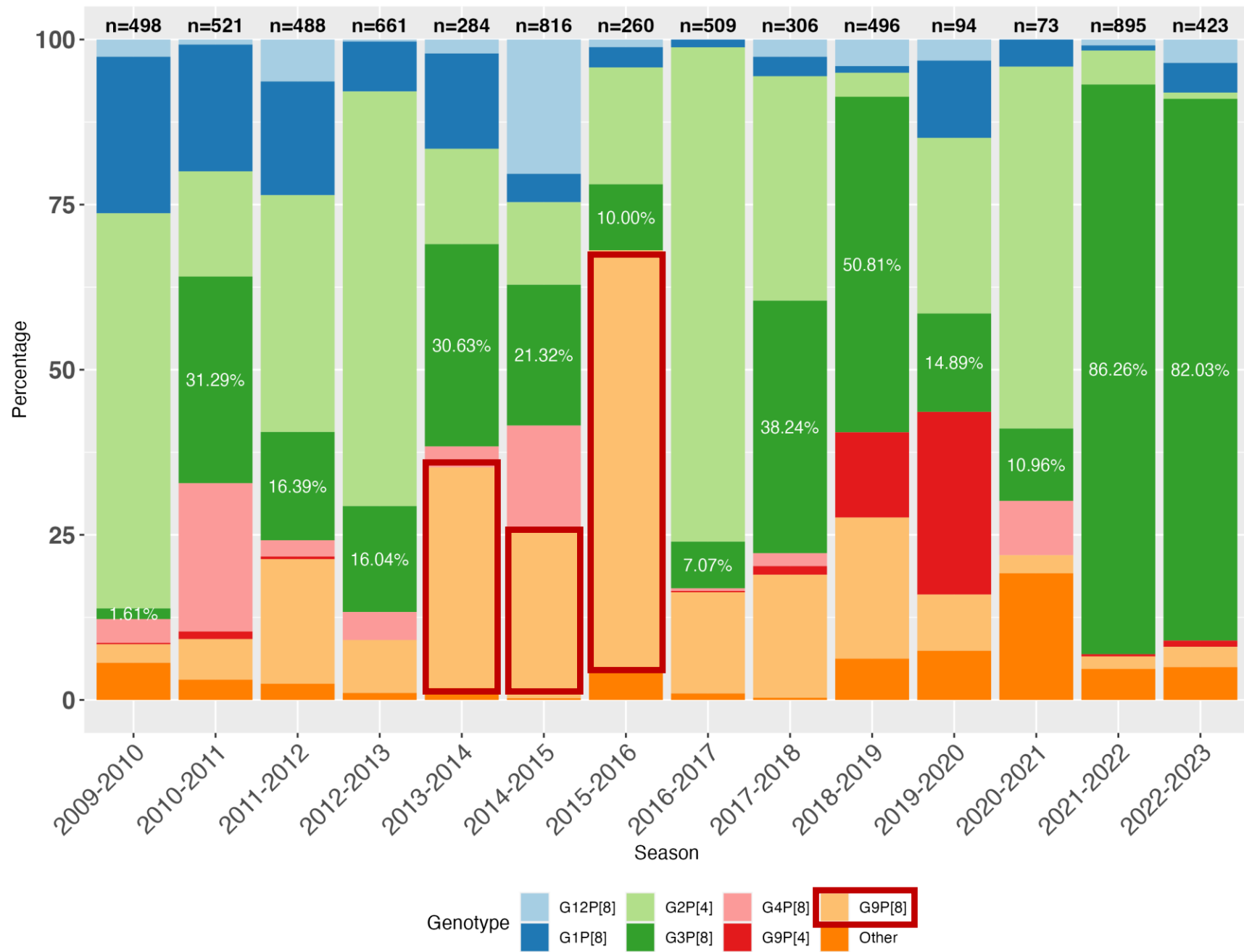


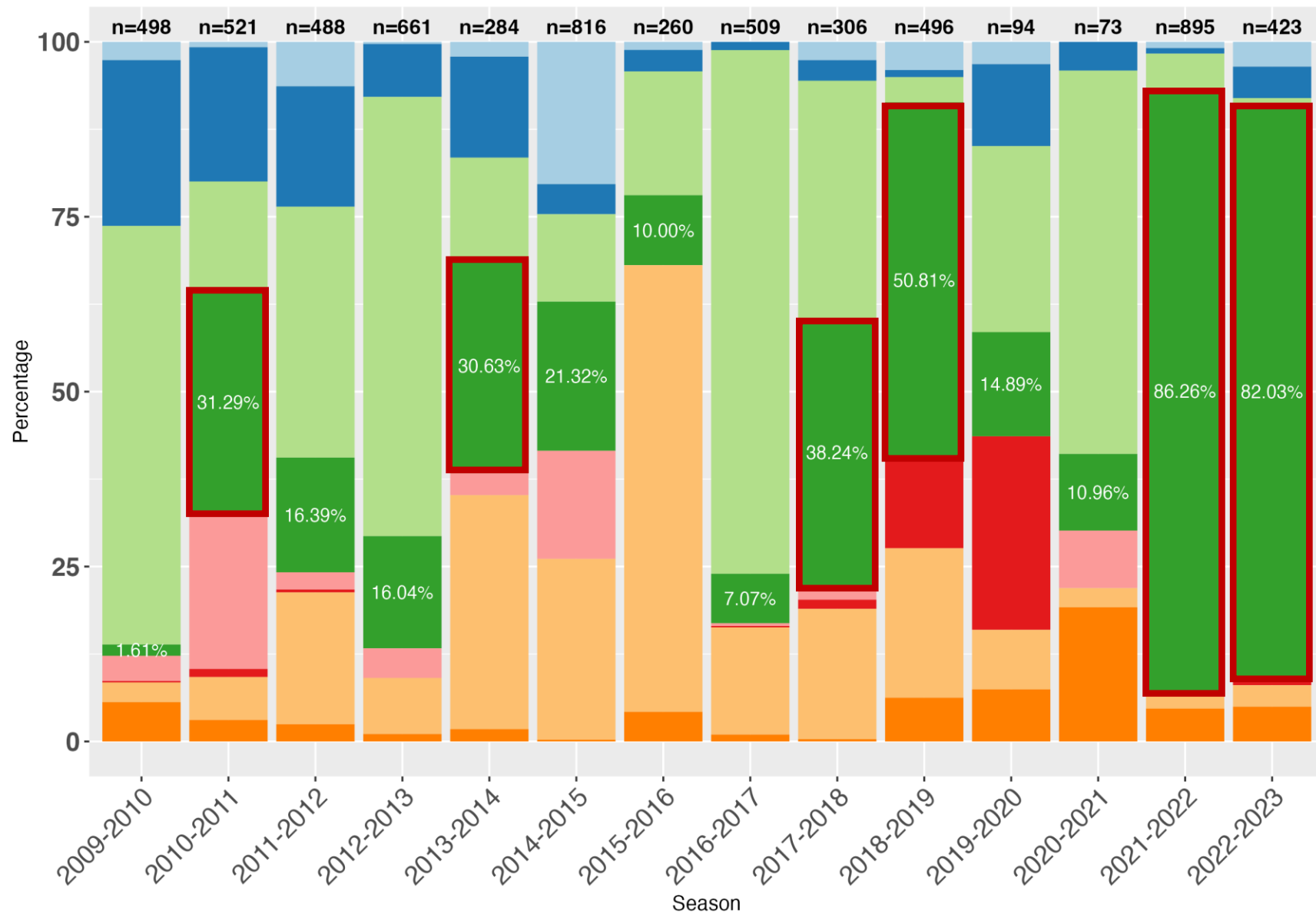
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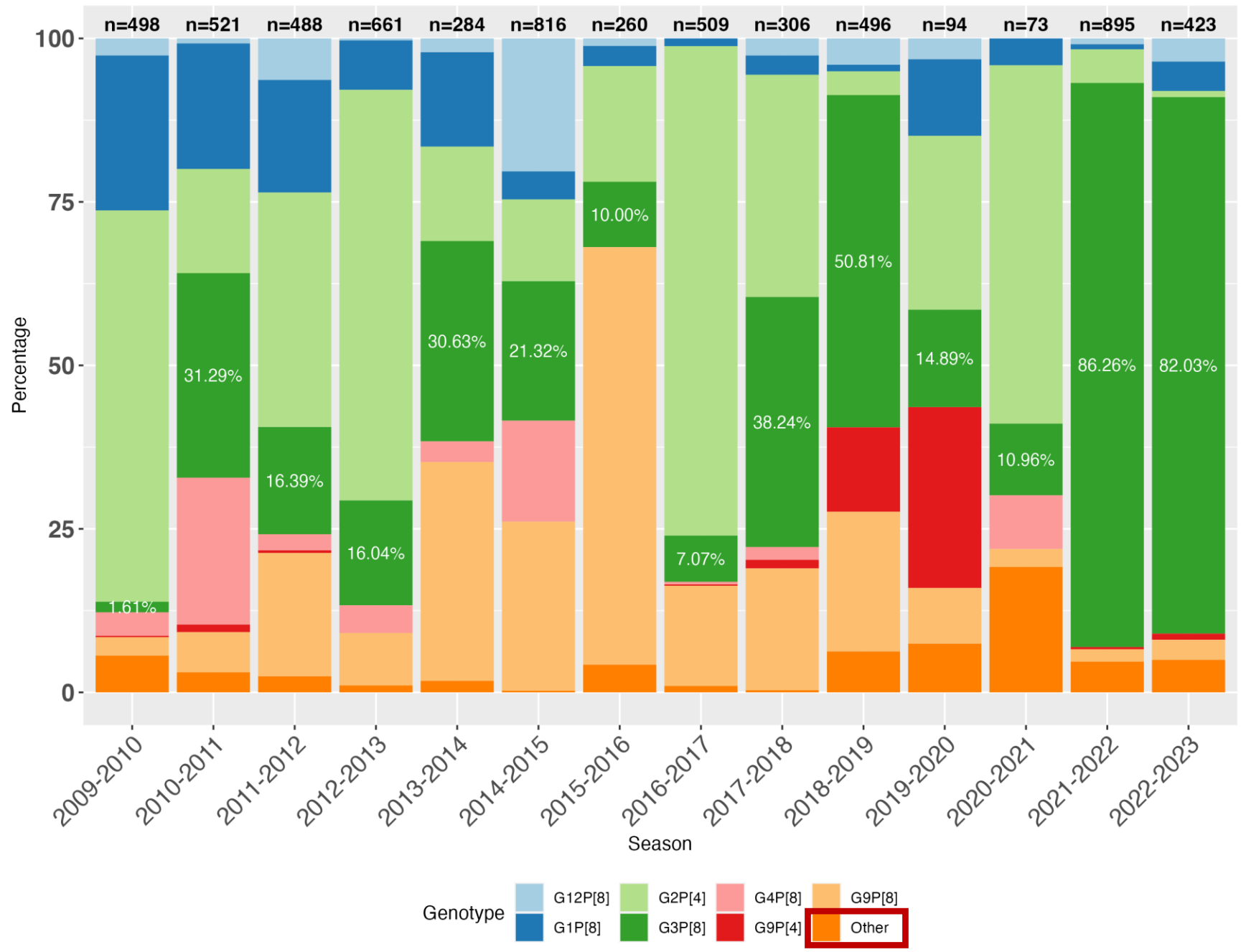








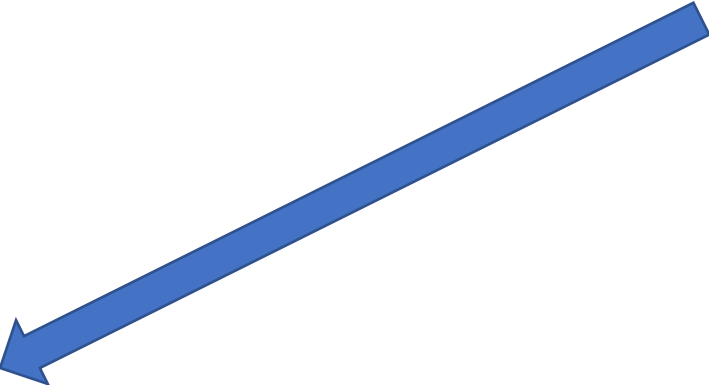






# Zoonosis

# Zoonosis



G3P[3]  
G3P[9]

1x  
1x

# Zoonosis



G3P[3]

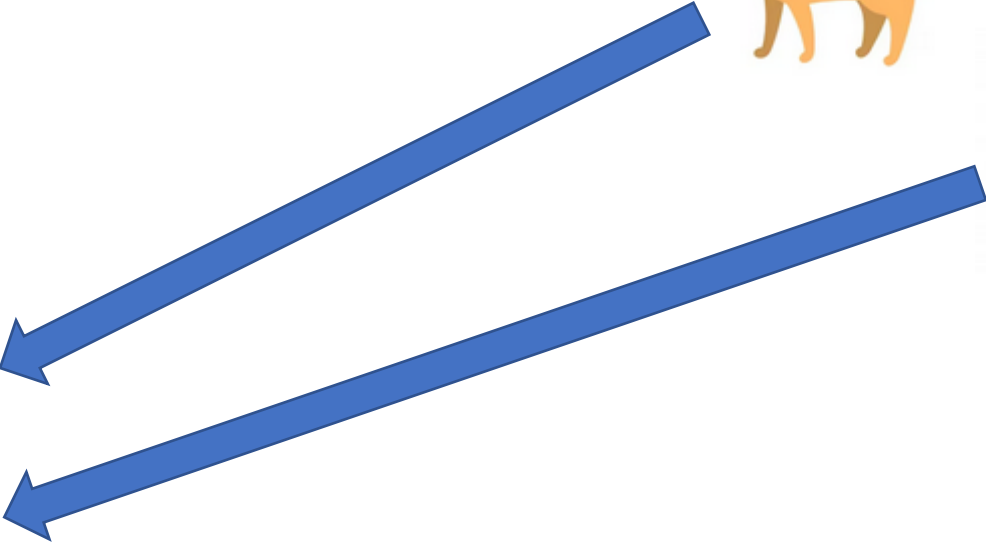
1x

G3P[9]

1x

G3P[14]

3x



# Zoonosis

G3P[3]

1x

G3P[9]

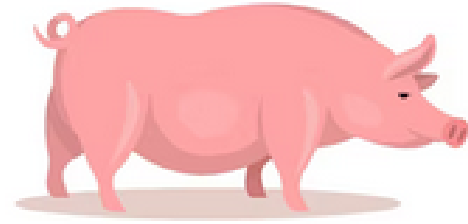
1x

G3P[14]

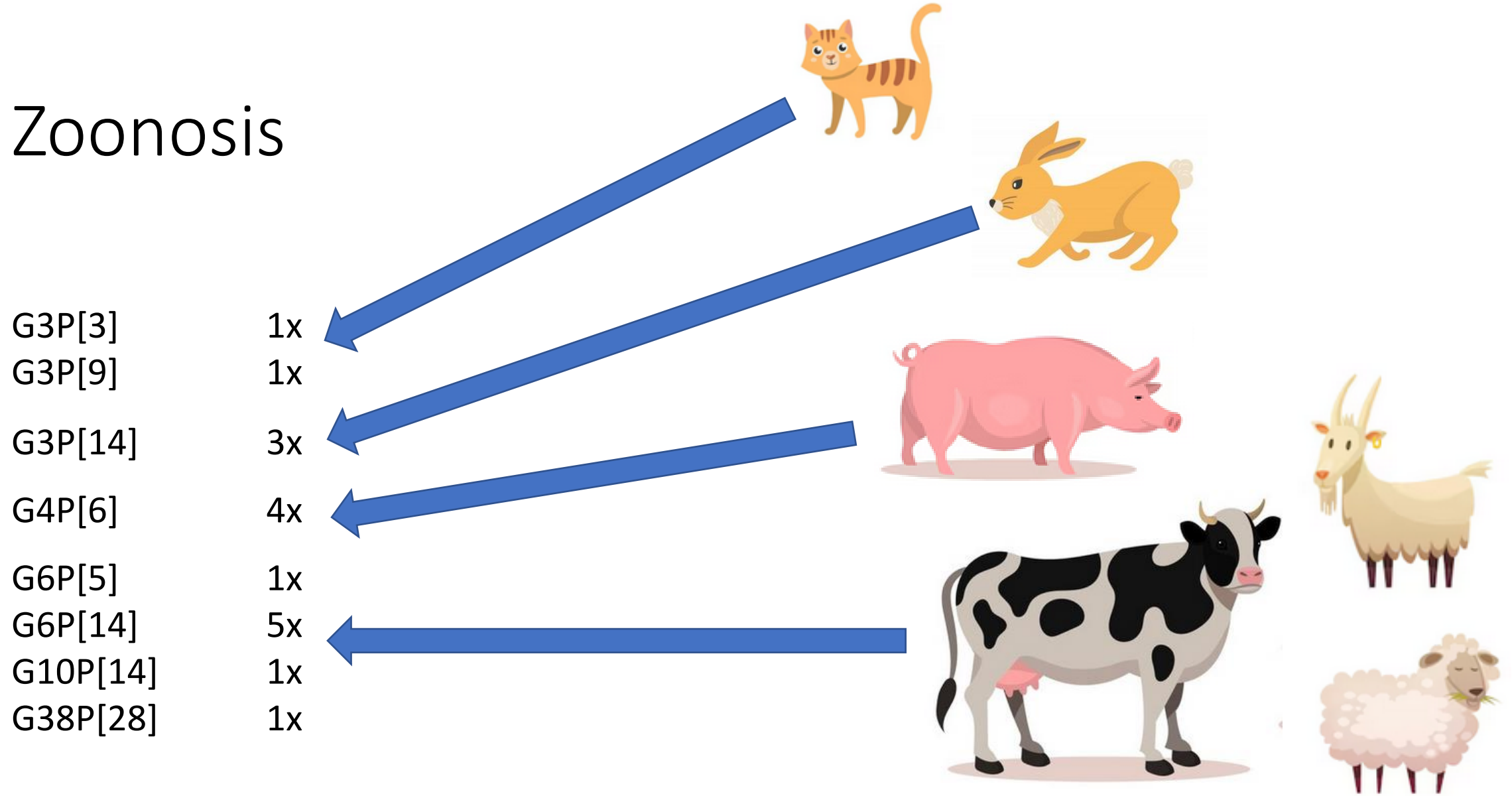
3x

G4P[6]

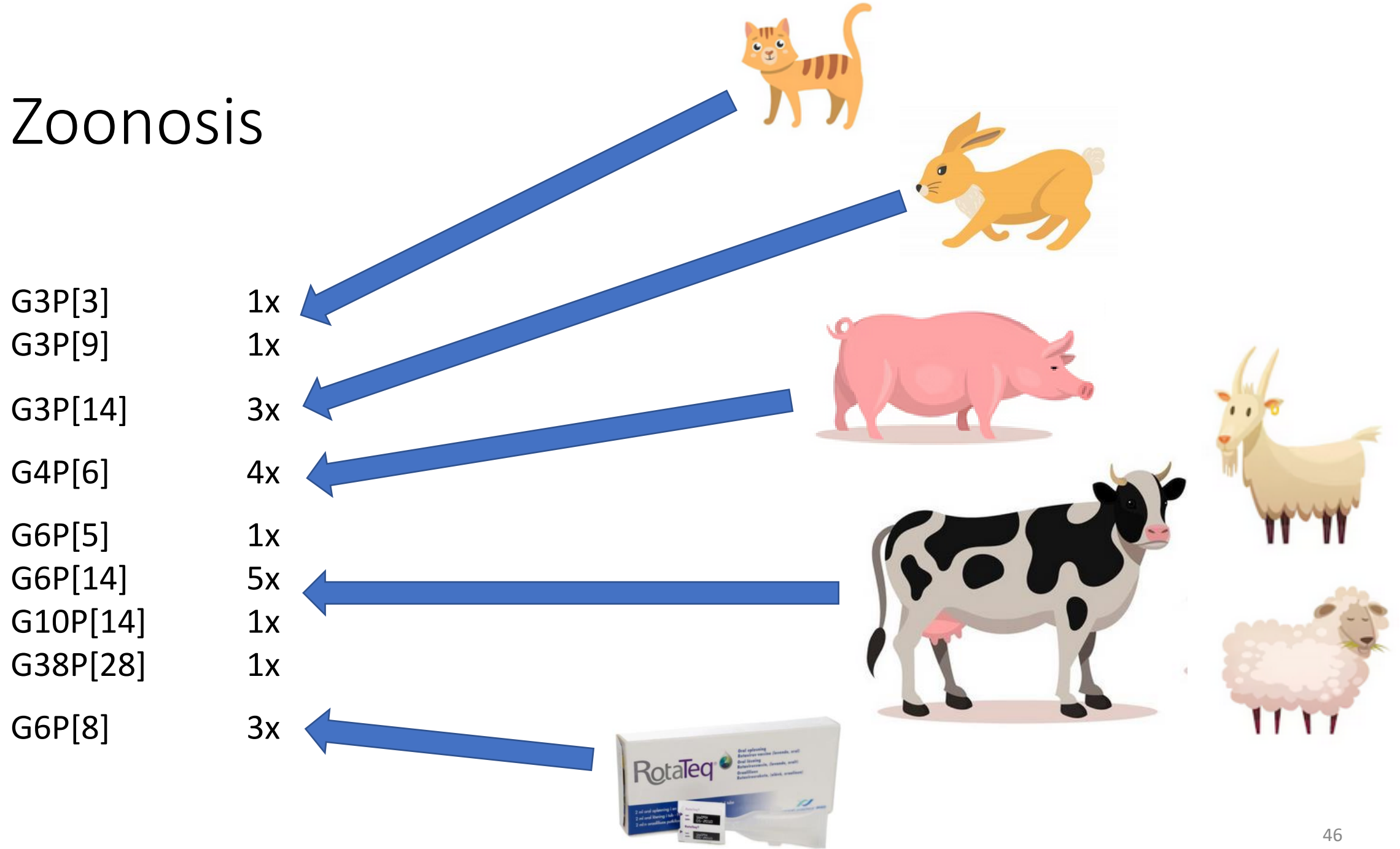
4x



# Zoonosis



# Zoonosis



# Rotavirus co-infections






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MICROBIOLOGY

Journal of  
Clinical Microbiology®

EPIDEMIOLOGY

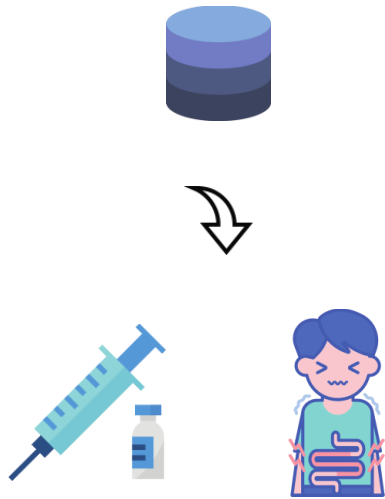
December 2021 Volume 59 Issue 12 10.1128/jcm.01236-21  
<https://doi.org/10.1128/jcm.01236-21>

# High Prevalence of Coinfecting Enteropathogens in Suspected Rotavirus Vaccine Breakthrough Cases

Ceren Simsek <sup>a</sup>, Mandy Bloemen<sup>a</sup>, Daan Jansen <sup>a</sup>, Leen Beller<sup>a</sup>, Patrick Descheemaeker<sup>b</sup>,  
Marijke Reynders<sup>b</sup>, Marc Van Ranst<sup>a</sup>, Jelle Matthijssens <sup>a</sup>

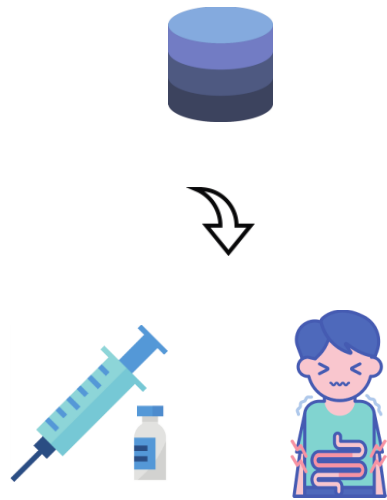


## NRC patient database

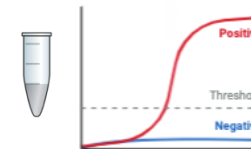


**102** samples  
from **2007-2008** to **2017-2018**

# NRC patient database



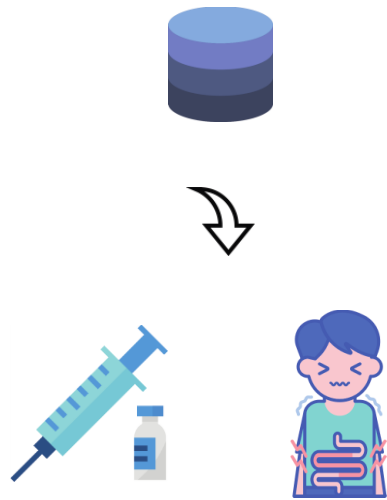
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RT-qPCR



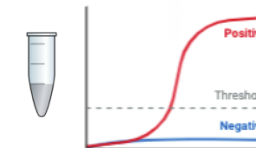
# NRC patient database



**102** samples  
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NGS



RT-qPCR



# NetoVIR protocol

# NetoVIR protocol

## SCIENTIFIC REPORTS



OPEN

**Modular approach to customise sample preparation procedures for viral metagenomics: a reproducible protocol for virome analysis**

Received: 19 August 2015

Accepted: 15 October 2015

Published: 12 November 2015

Nádia Conceição-Neto<sup>1,2</sup>, Mark Zeller<sup>1</sup>, Hanne Lefrère<sup>1</sup>, Pieter De Bruyn<sup>1</sup>, Leen Beller<sup>1</sup>, Ward Deboutte<sup>1</sup>, Claude Kwe Yinda<sup>1,2</sup>, Rob Lavigne<sup>3</sup>, Piet Maes<sup>2</sup>, Marc Van Ranst<sup>2</sup>, Elisabeth Heylen<sup>1,\*</sup> & Jelle Matthijssens<sup>1,2,\*</sup>

# NetoVIR protocol

# SCIENTIFIC REPORTS

OPEN

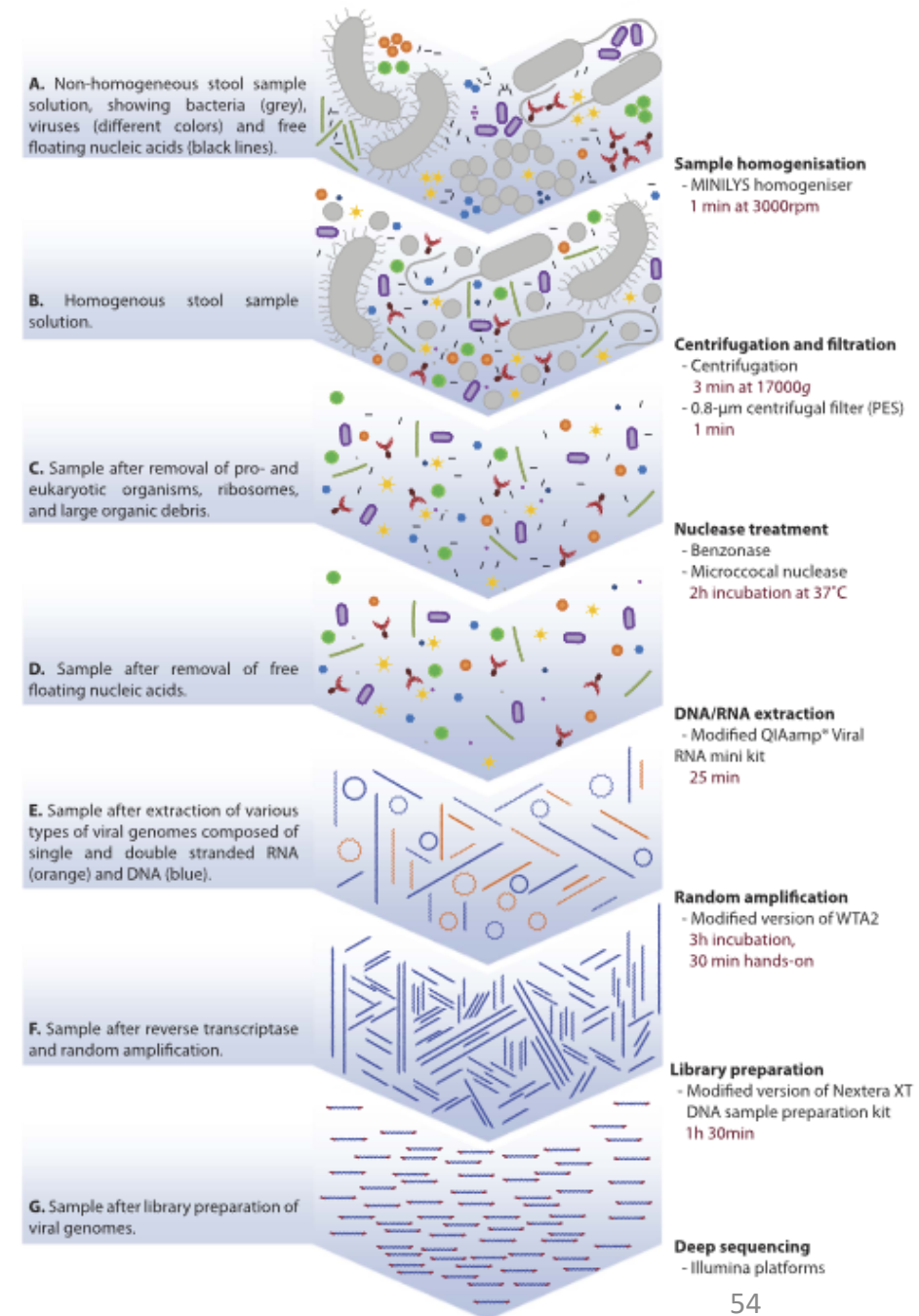
## Modular approach to customise sample preparation procedures for viral metagenomics: a reproducible protocol for virome analysis

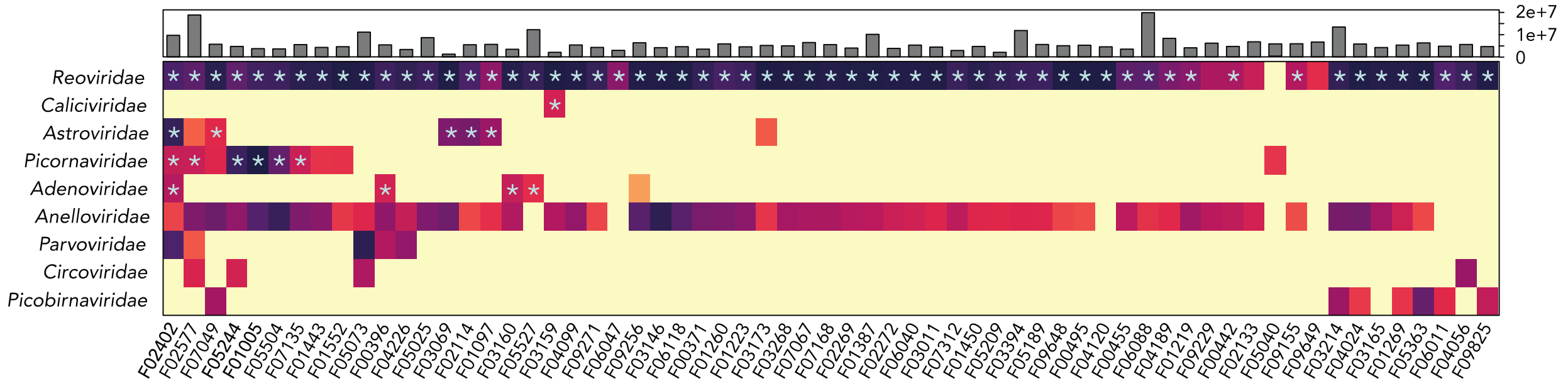
Received: 19 August 2015

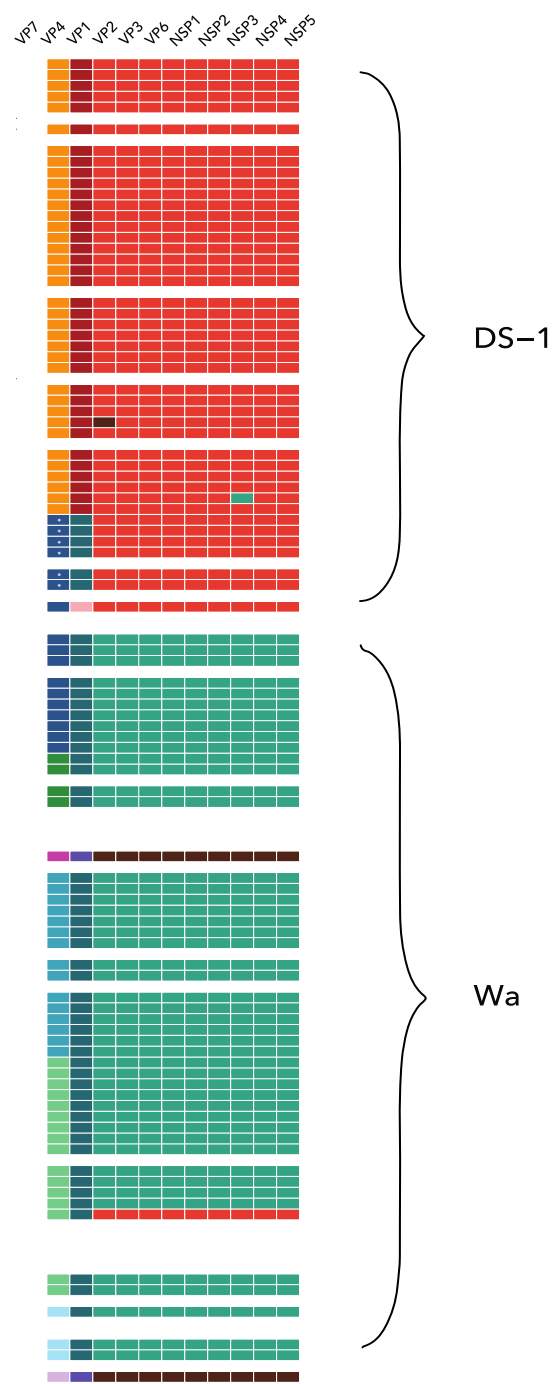
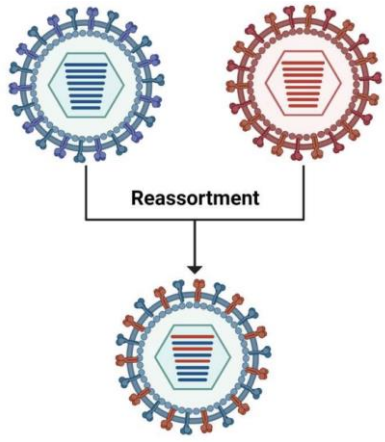
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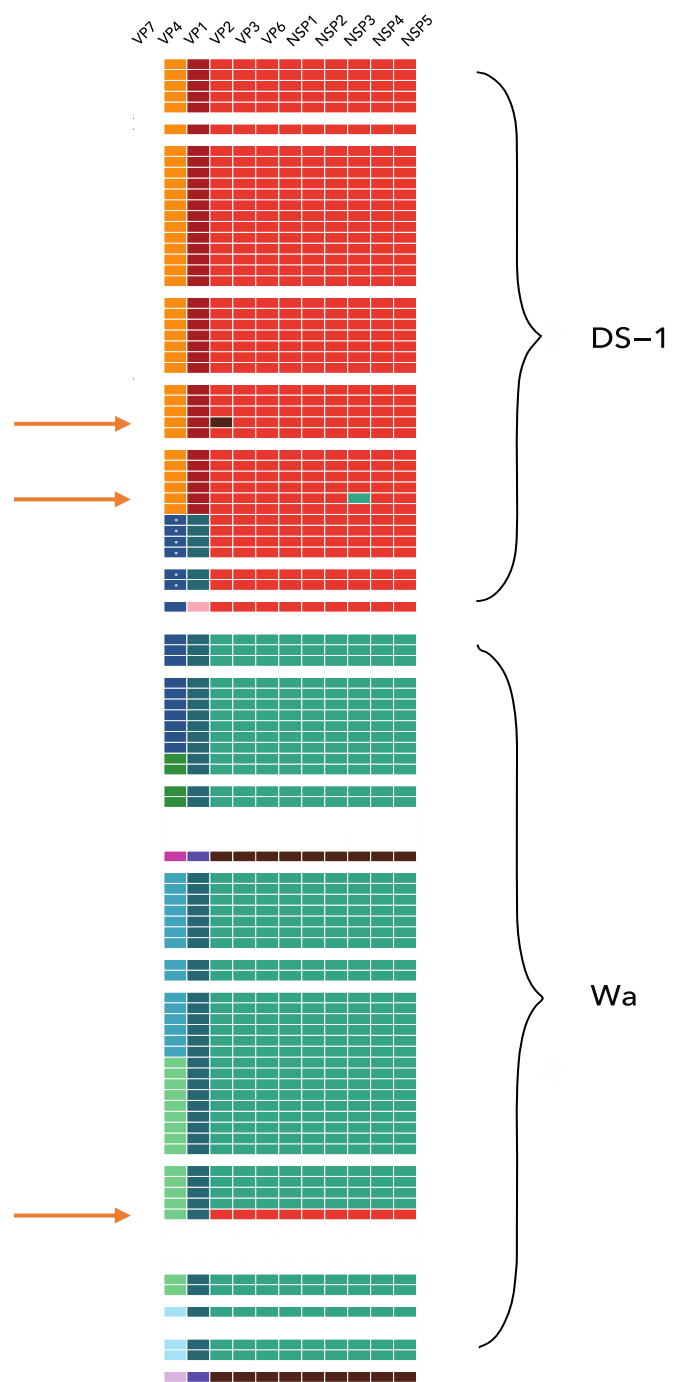
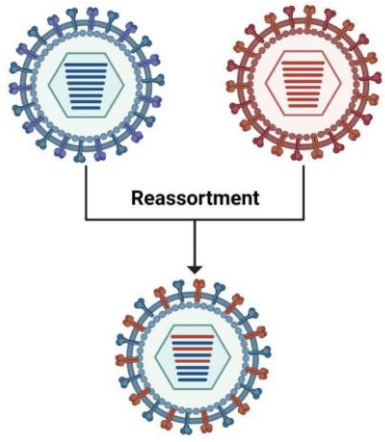
Nádia Conceição-Neto<sup>1,2</sup>, Mark Zeller<sup>1</sup>, Hanne Lefrère<sup>1</sup>, Pieter De Bruyn<sup>1</sup>, Leen Beller<sup>1</sup>, Ward Deboutte<sup>1</sup>, Claude Kwe Yinda<sup>1,2</sup>, Rob Lavigne<sup>3</sup>, Piet Maes<sup>2</sup>, Marc Van Ranst<sup>2</sup>, Elisabeth Heylen<sup>1,\*</sup> & Jelle Matthijssens<sup>1,2,\*</sup>



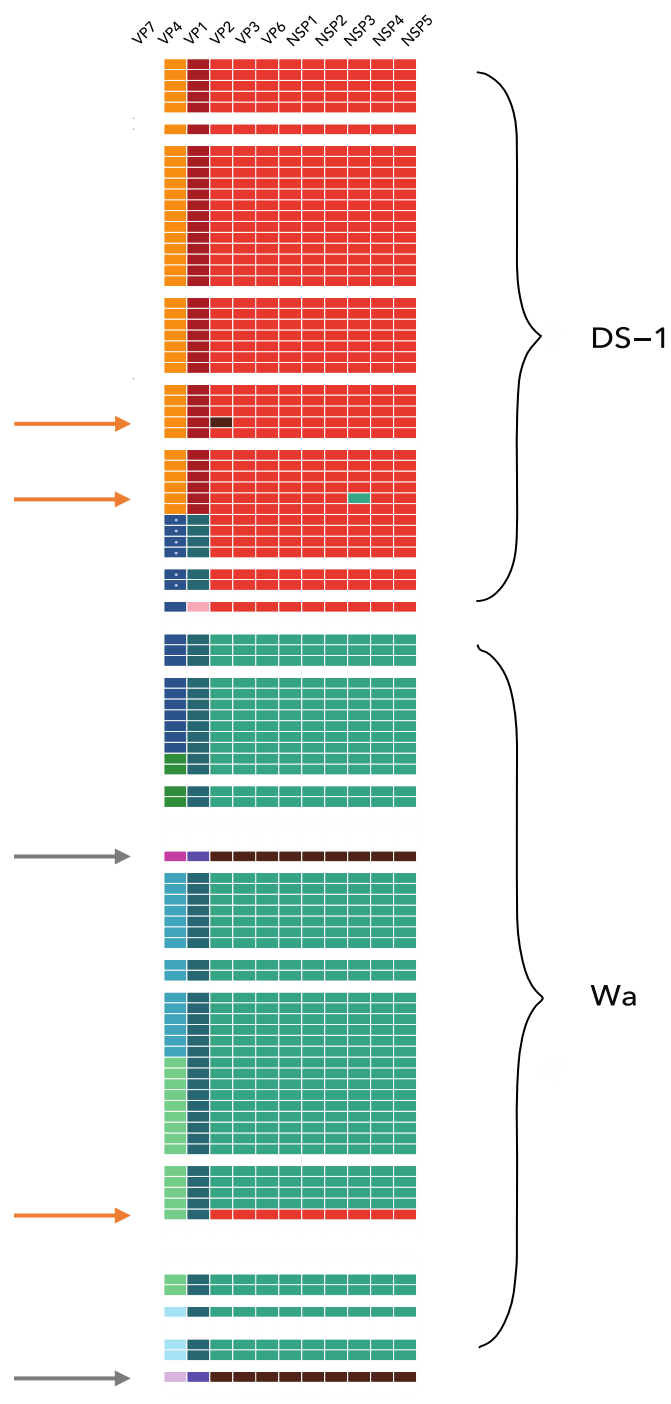
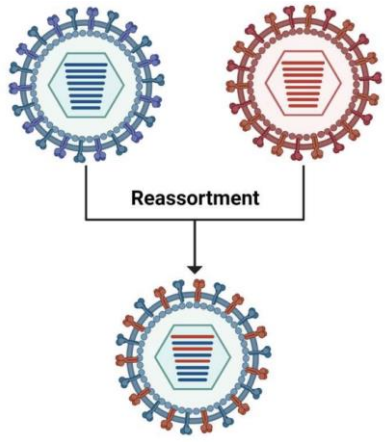




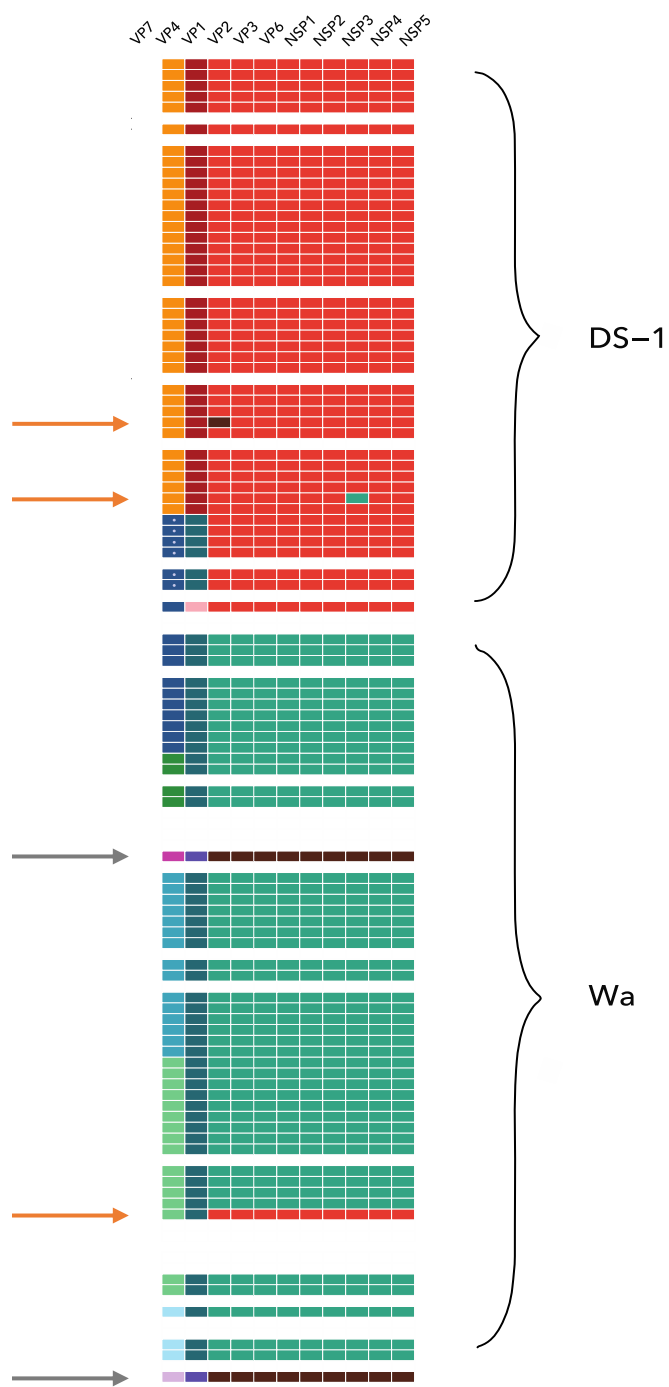
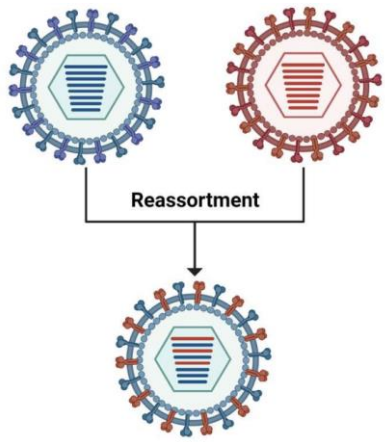




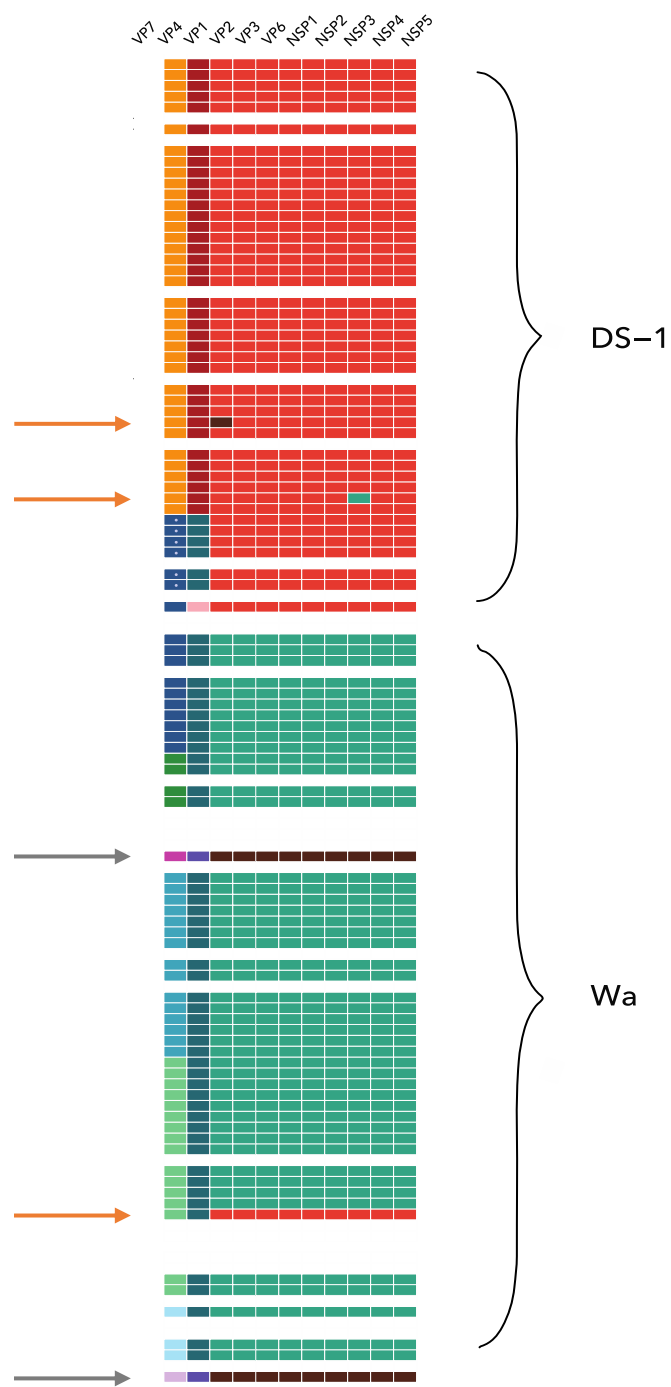
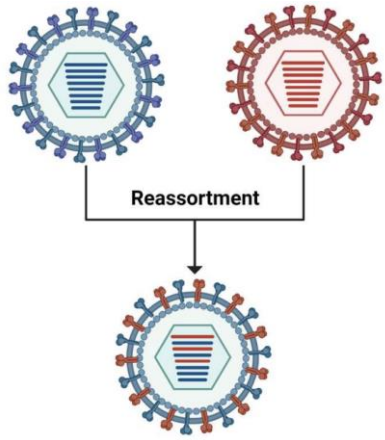
○ Few intergenogroup reassortant



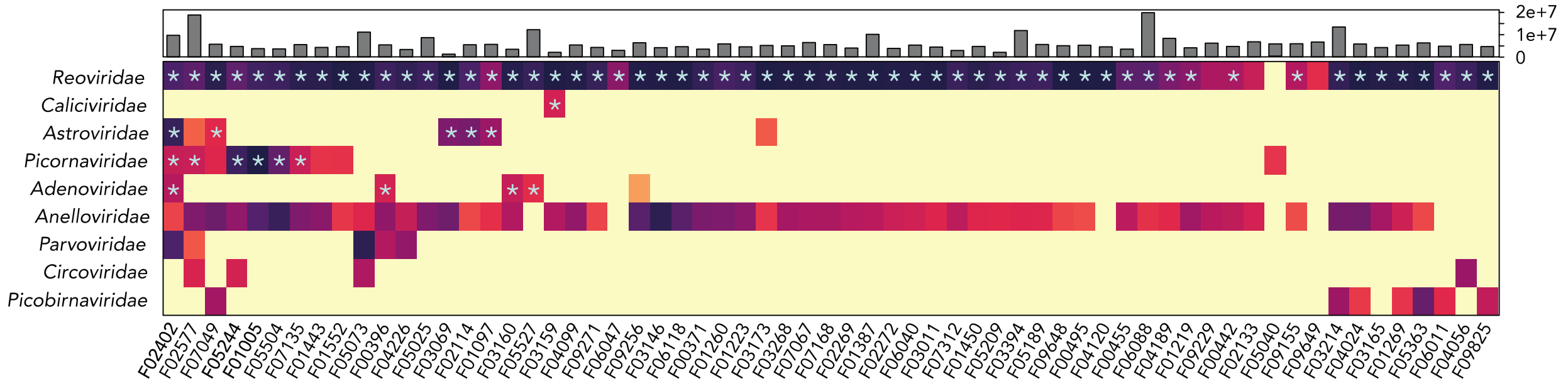
- Few intergenogroup reassortant
- Few zoonotic strains detected



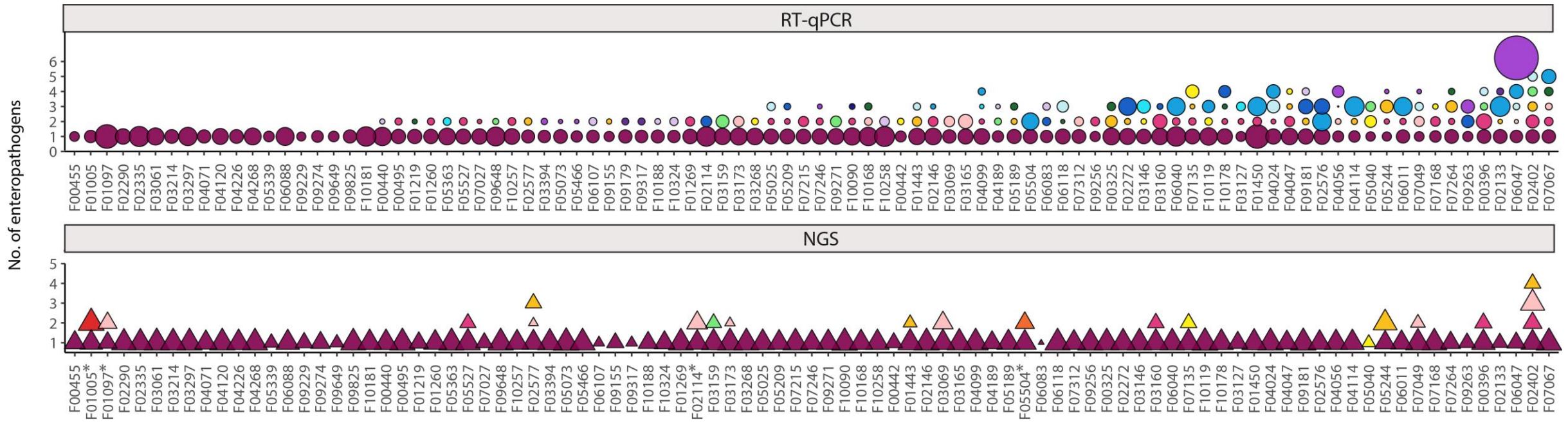
- Few intergenogroup reassortant
- Few zoonotic strains detected
- → Fitness cost

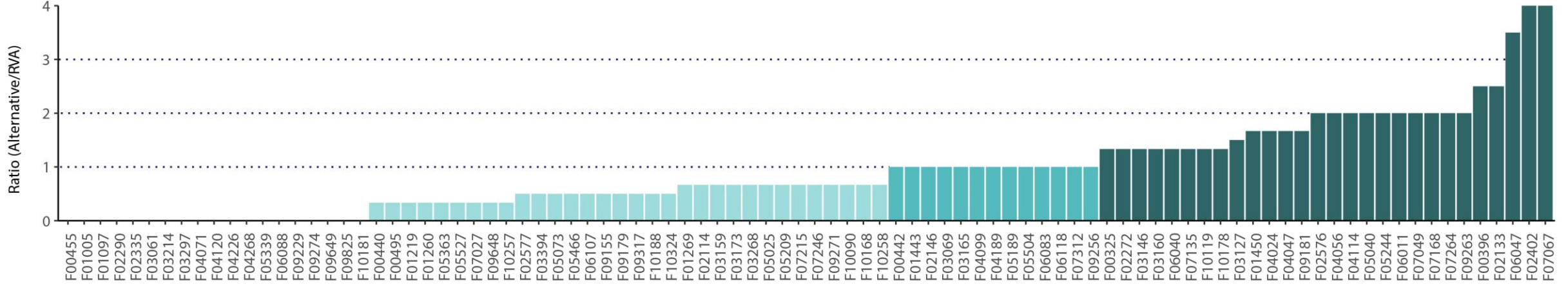
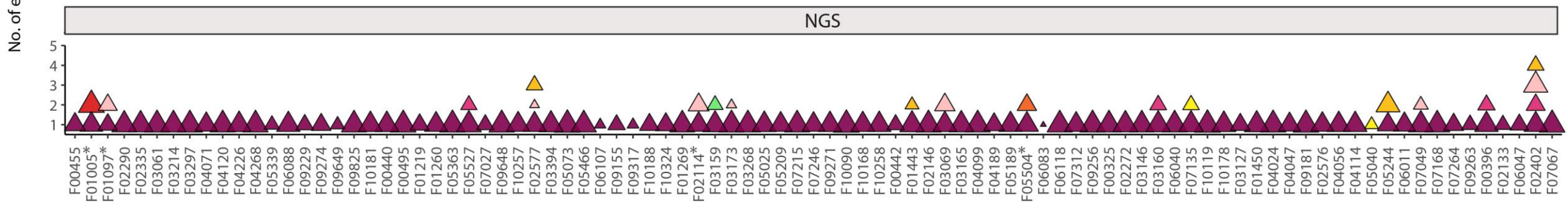
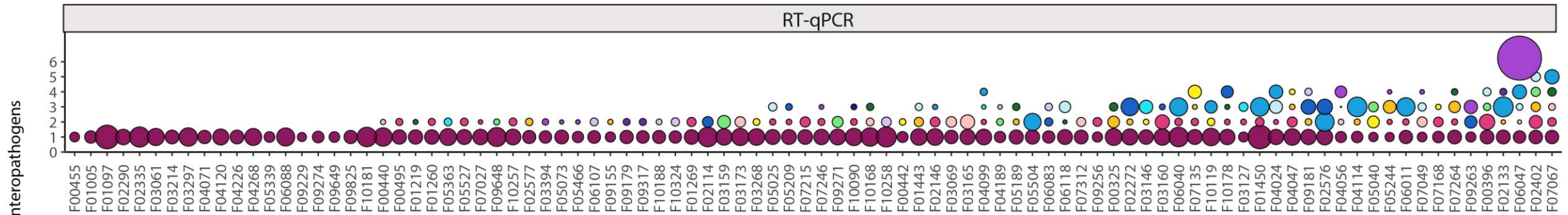


- Few intergenogroup reassortant
- Few zoonotic strains detected
- → Fitness cost
- **No** evidence for unusual genotypes that might suggest escape vaccine-induced immunity









**Breakthrough**

**Equal**

**Alternative**



# Summary

- No unusual rotavirus genotypes in breakthrough cases
  - ✓ Few reassortants and few inter species transmissions
- Various other viral, bacterial and parasitic co-infecting pathogens
- Approximately 50% of cases might be explained by other pathogens
- Implications for diagnostics and clinical trials

# Vaccine-derived rotavirus strains



ELSEVIER

## Vaccine

Volume 40, Issue 35, 19 August 2022, Pages 5114-5125



# Rotavirus vaccine-derived cases in Belgium: Evidence for reversion of attenuating mutations and alternative causes of gastroenteritis

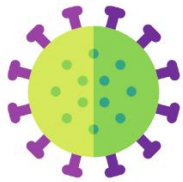
Ceren Simsek<sup>a</sup>, Mandy Bloemen<sup>a</sup>, Daan Jansen<sup>a</sup>, Patrick Descheemaeker<sup>b</sup>, Marijke Reynders<sup>b</sup>,

Marc Van Ranst<sup>a</sup>, Jelle Matthijnsens<sup>a</sup>  

## NRC patient database

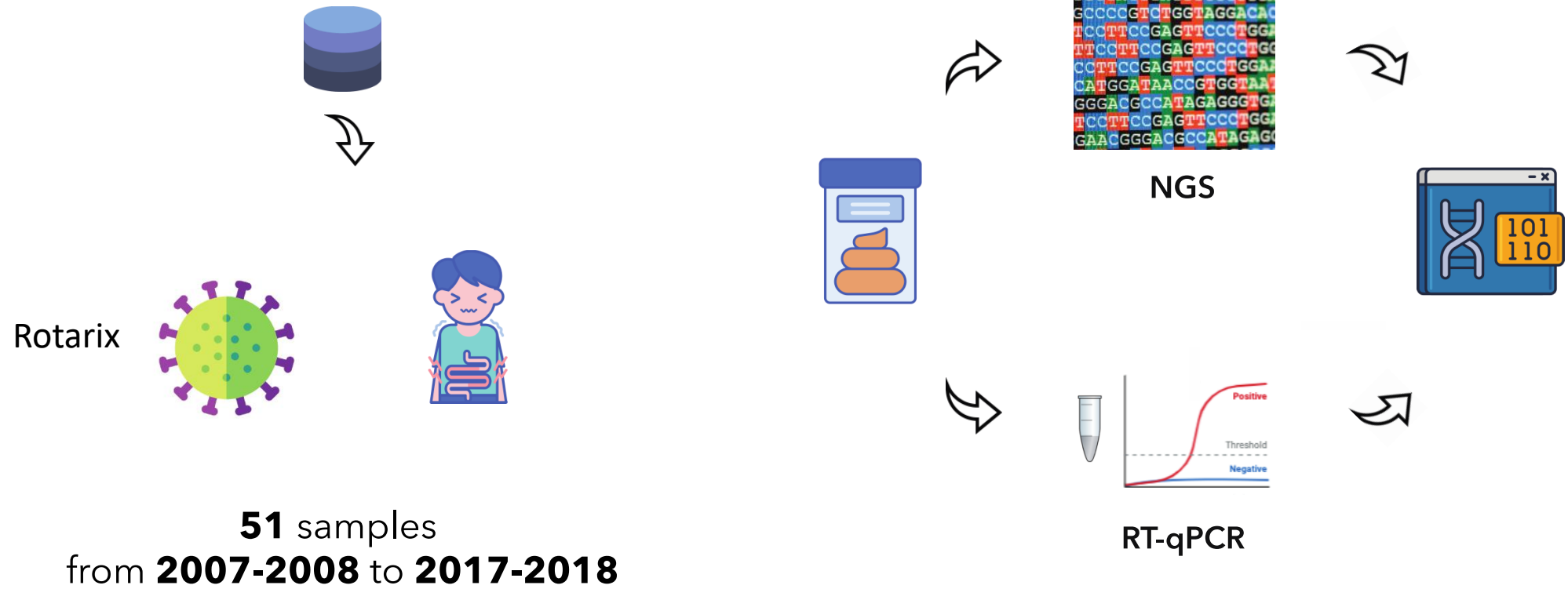


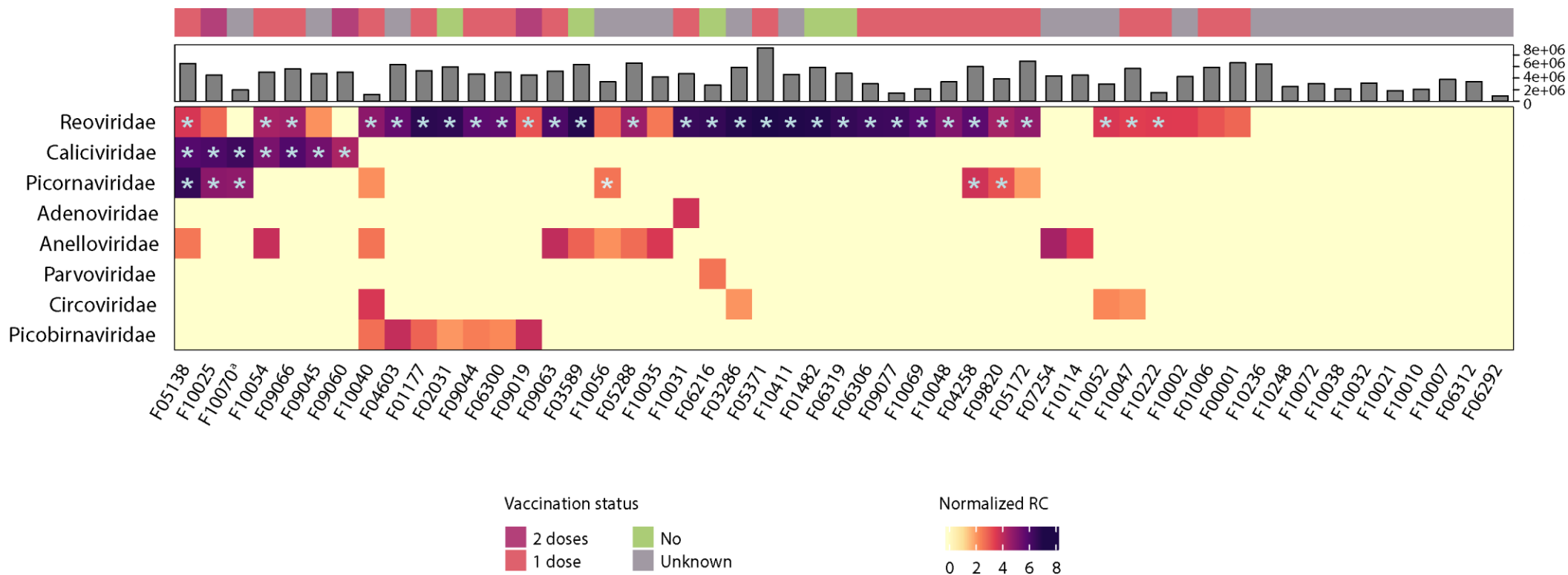
Rotarix

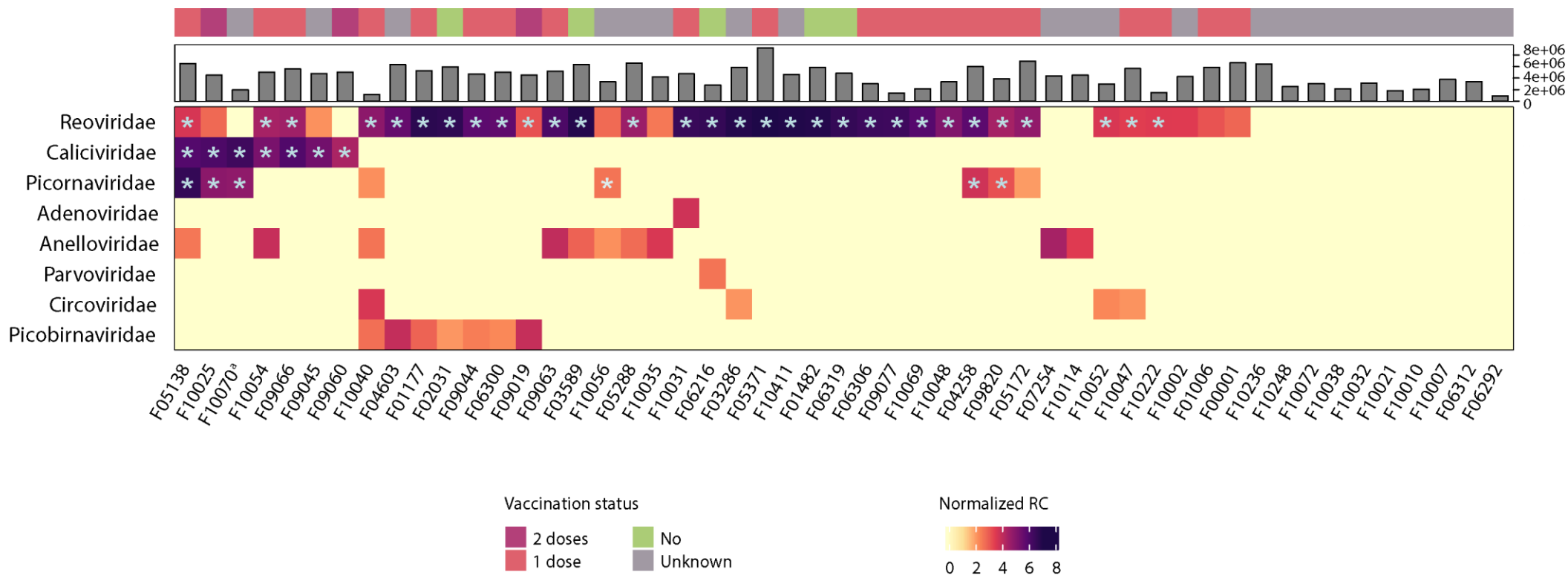


**51** samples  
from **2007-2008** to **2017-2018**

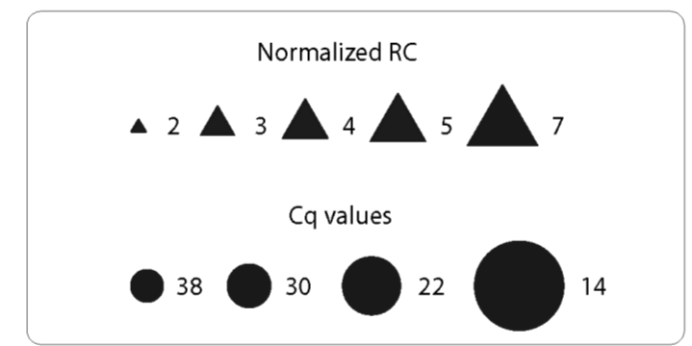
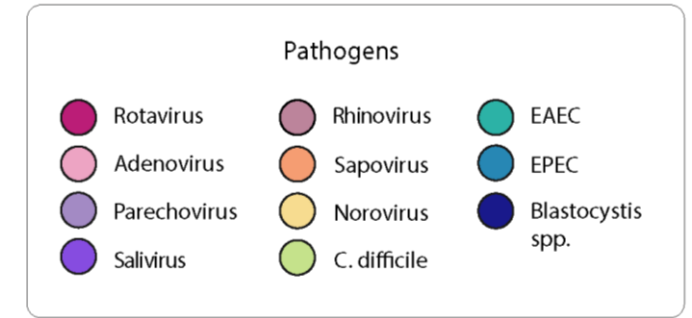
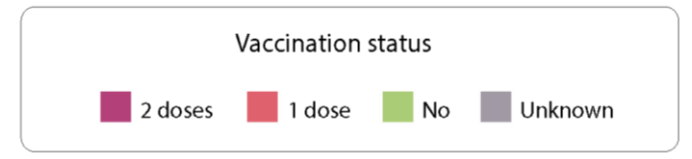
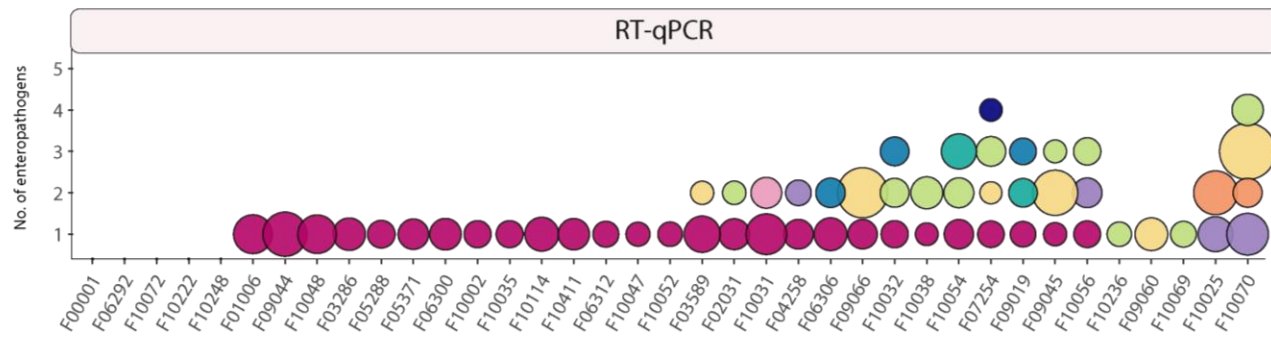
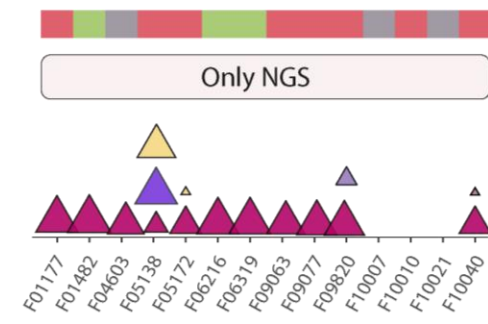
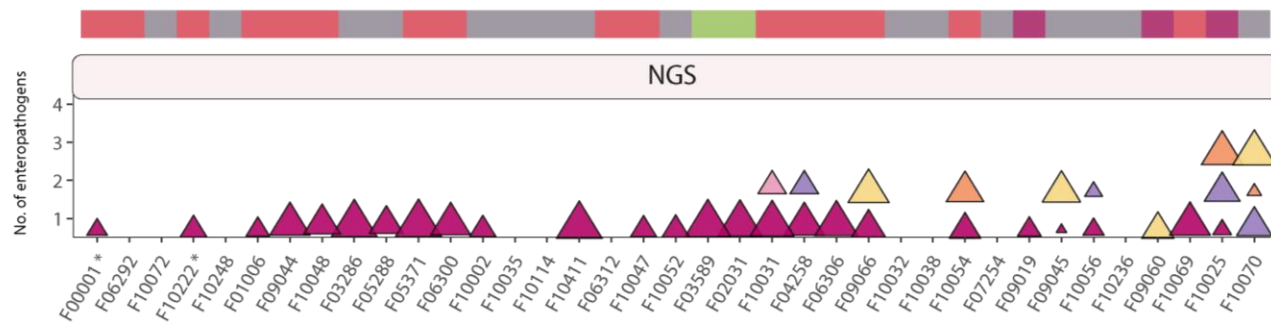
# NRC patient database



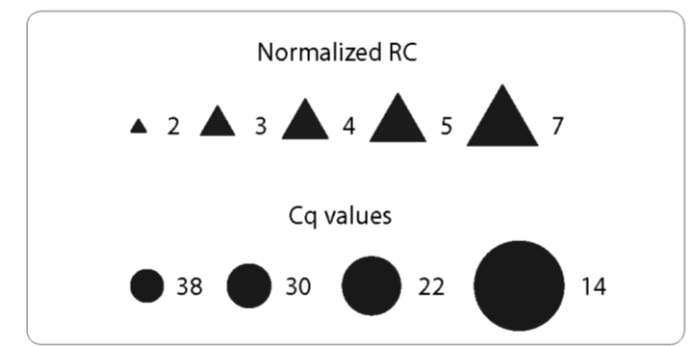
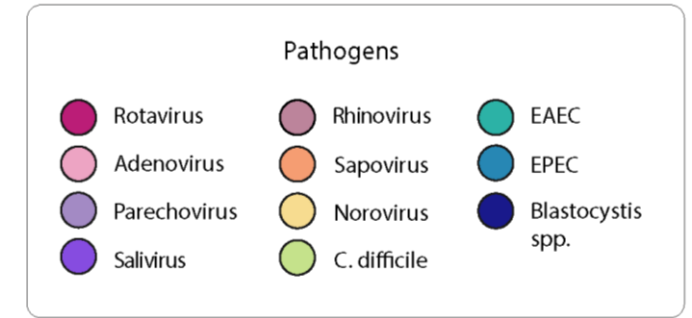
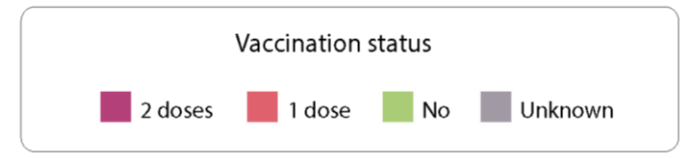
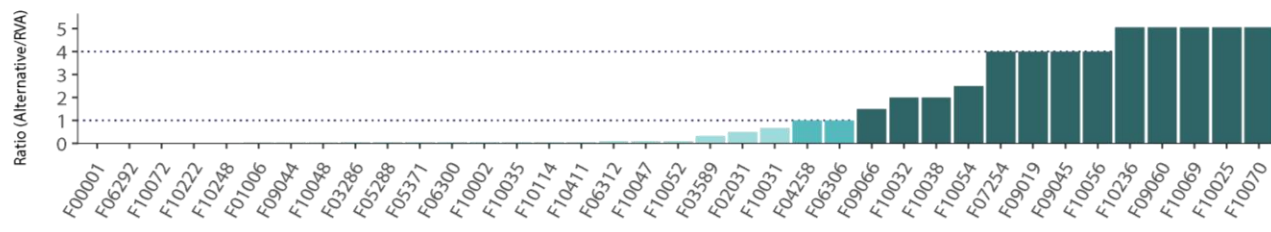
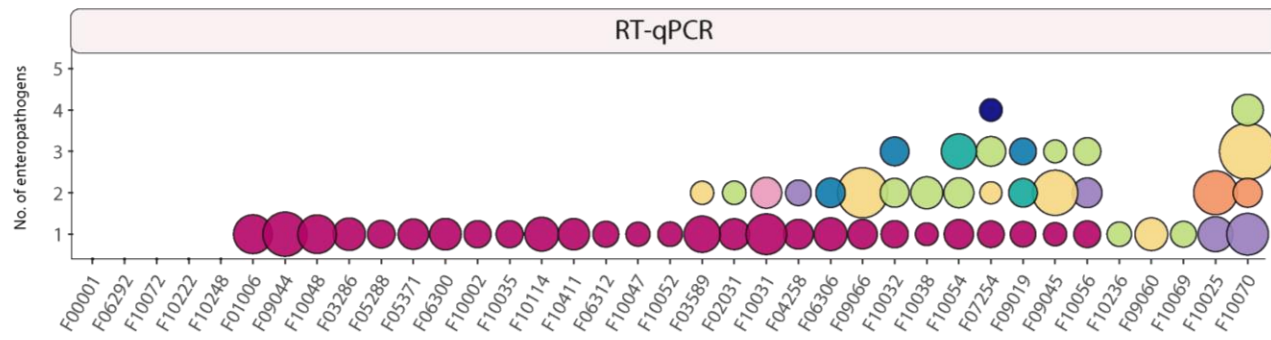
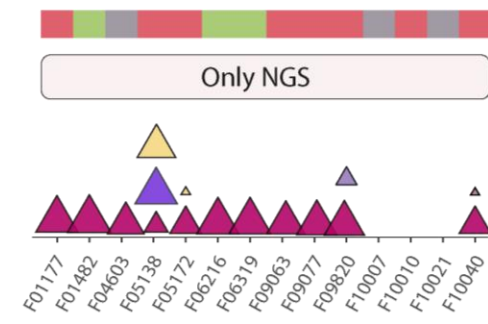
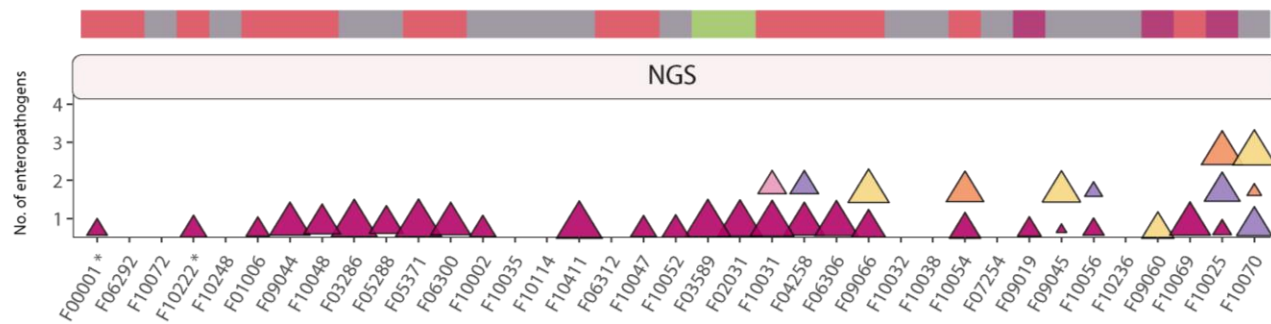




No evidence of reassortment between vaccine strains and wild type strains







# Origin of vaccine strains?

# Origin of vaccine strains?

Vaccine recipients shedding virus?



# Origin of vaccine strains?

Vaccine recipients shedding virus?

Transmission from vaccinated infant?



# Origin of vaccine strains?

Vaccine recipients shedding virus?

Transmission from vaccinated infant?

Circulating in the human population?

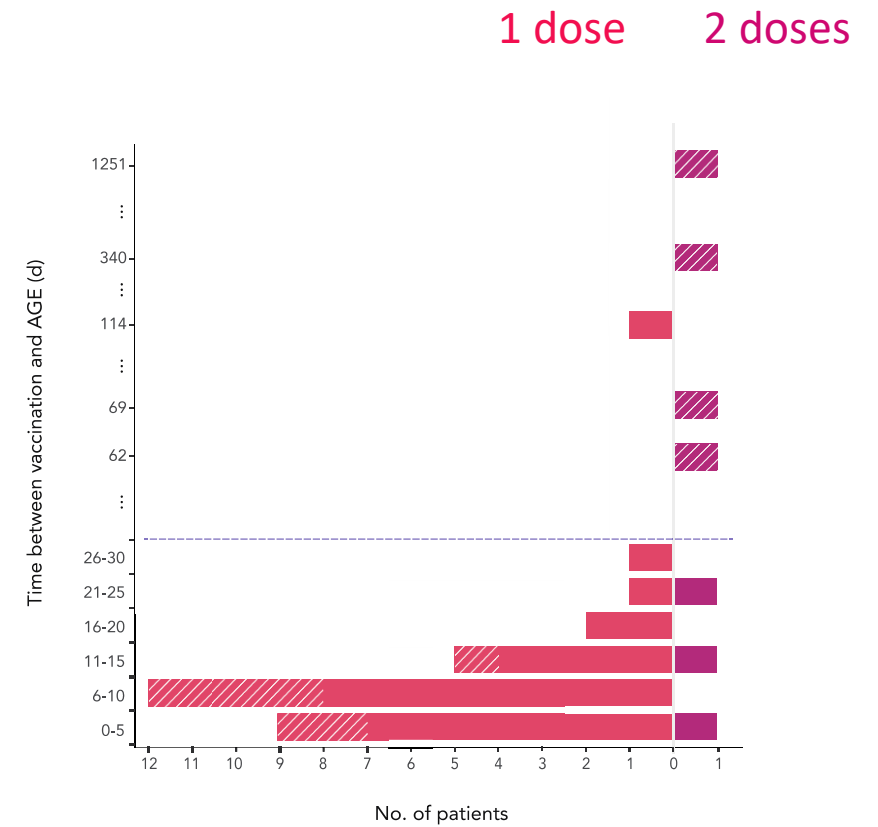


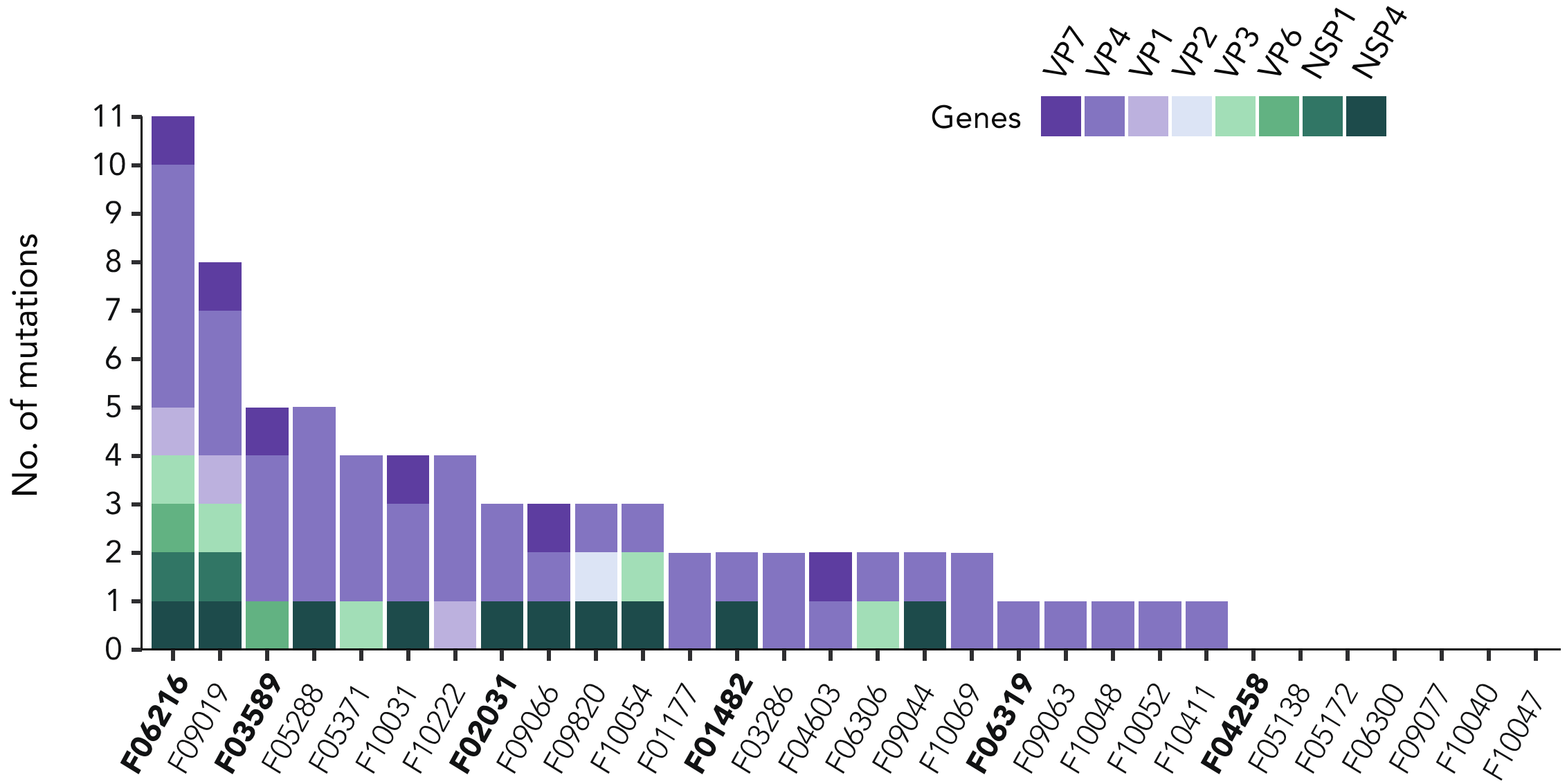
# Origin of vaccine strains?

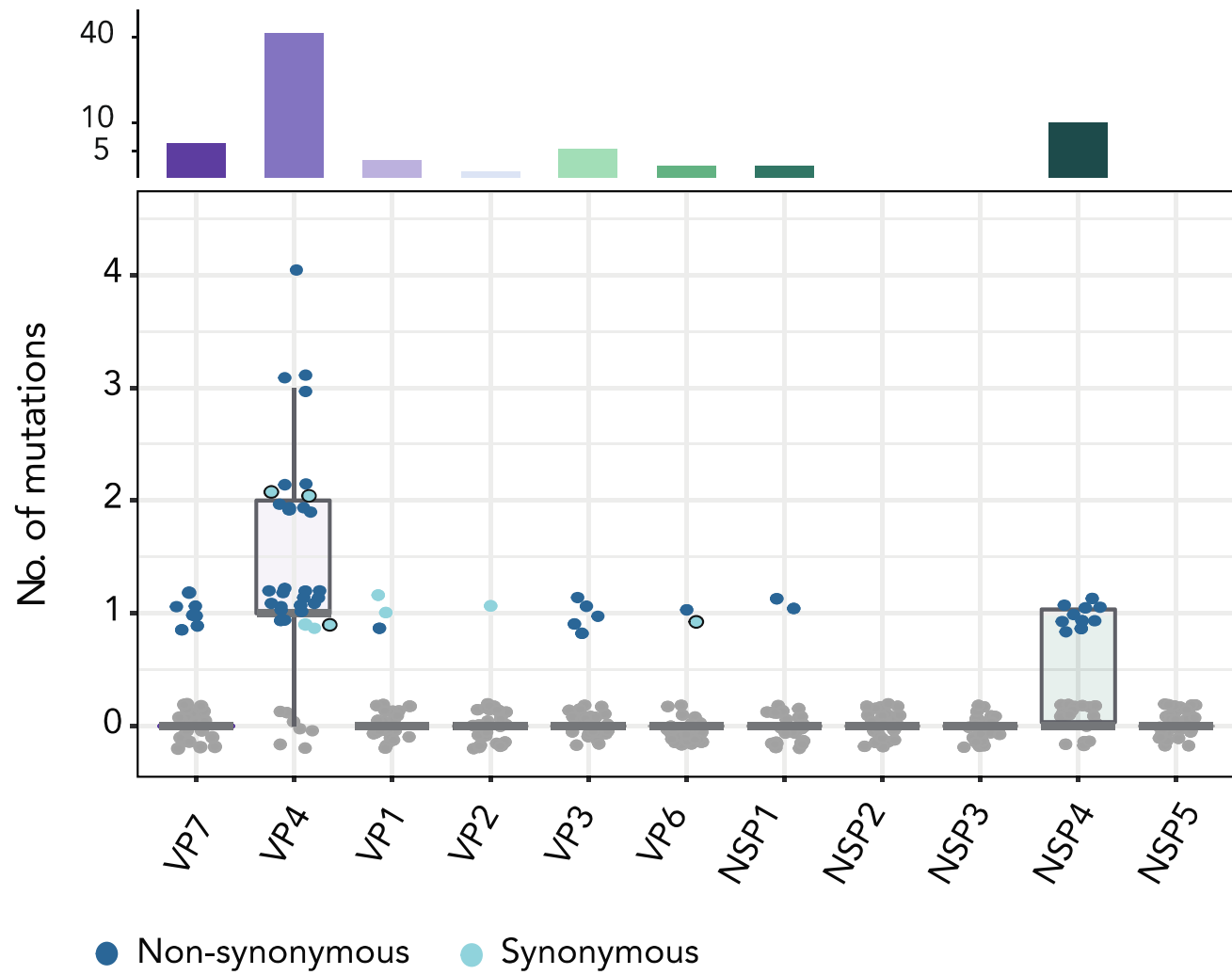
Vaccine recipients shedding virus?

Transmission from vaccinated infant?

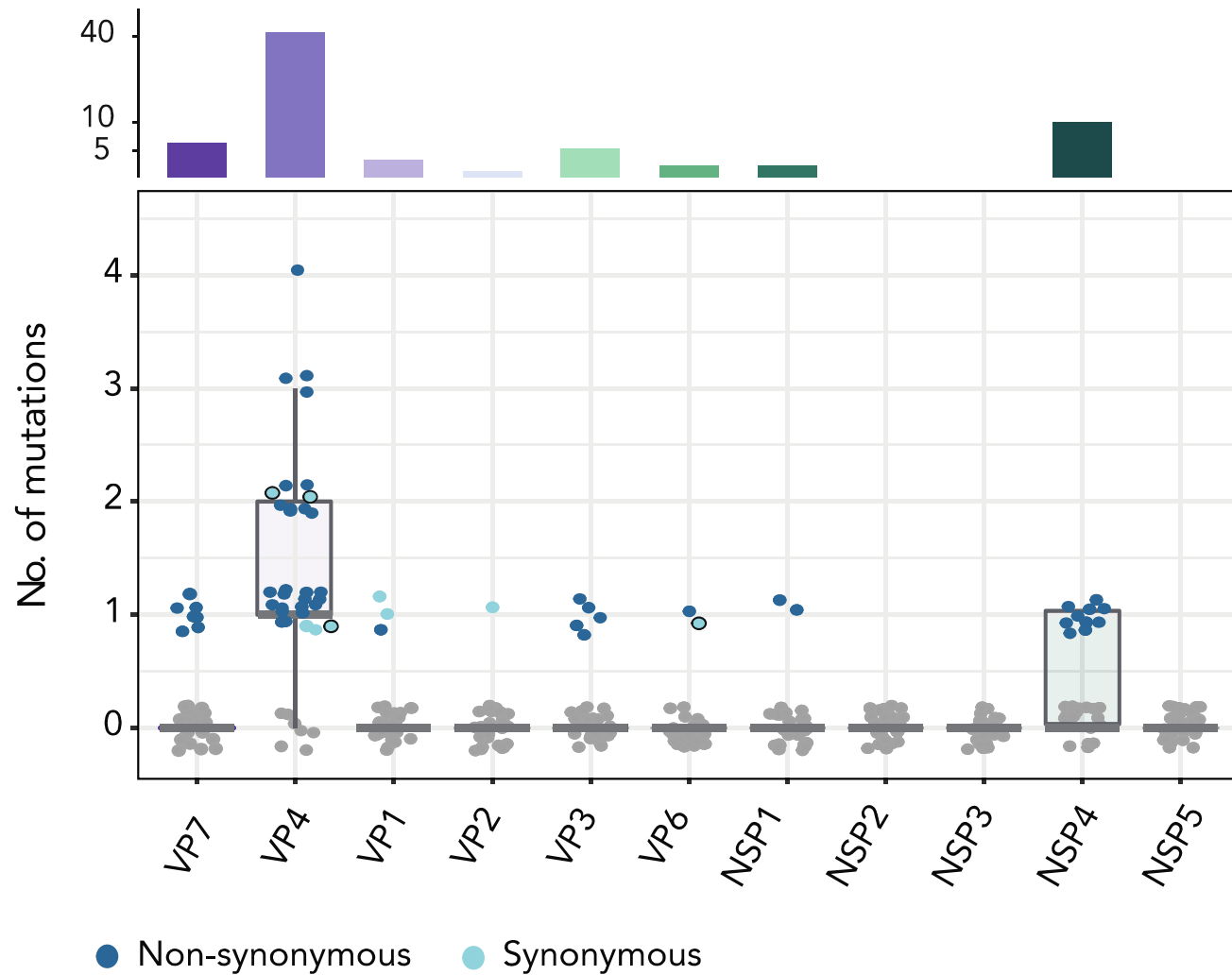
Circulating in the human population?



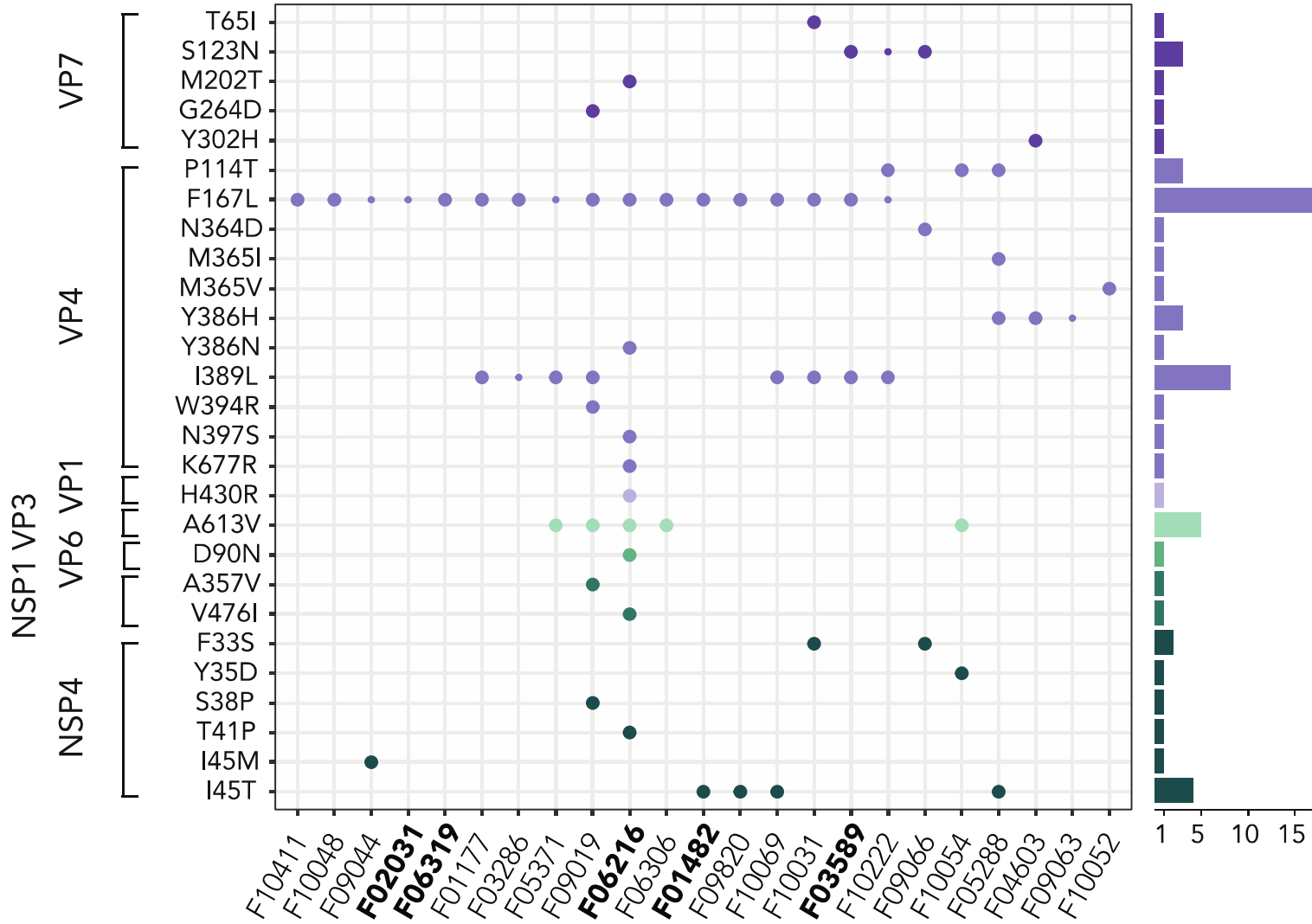


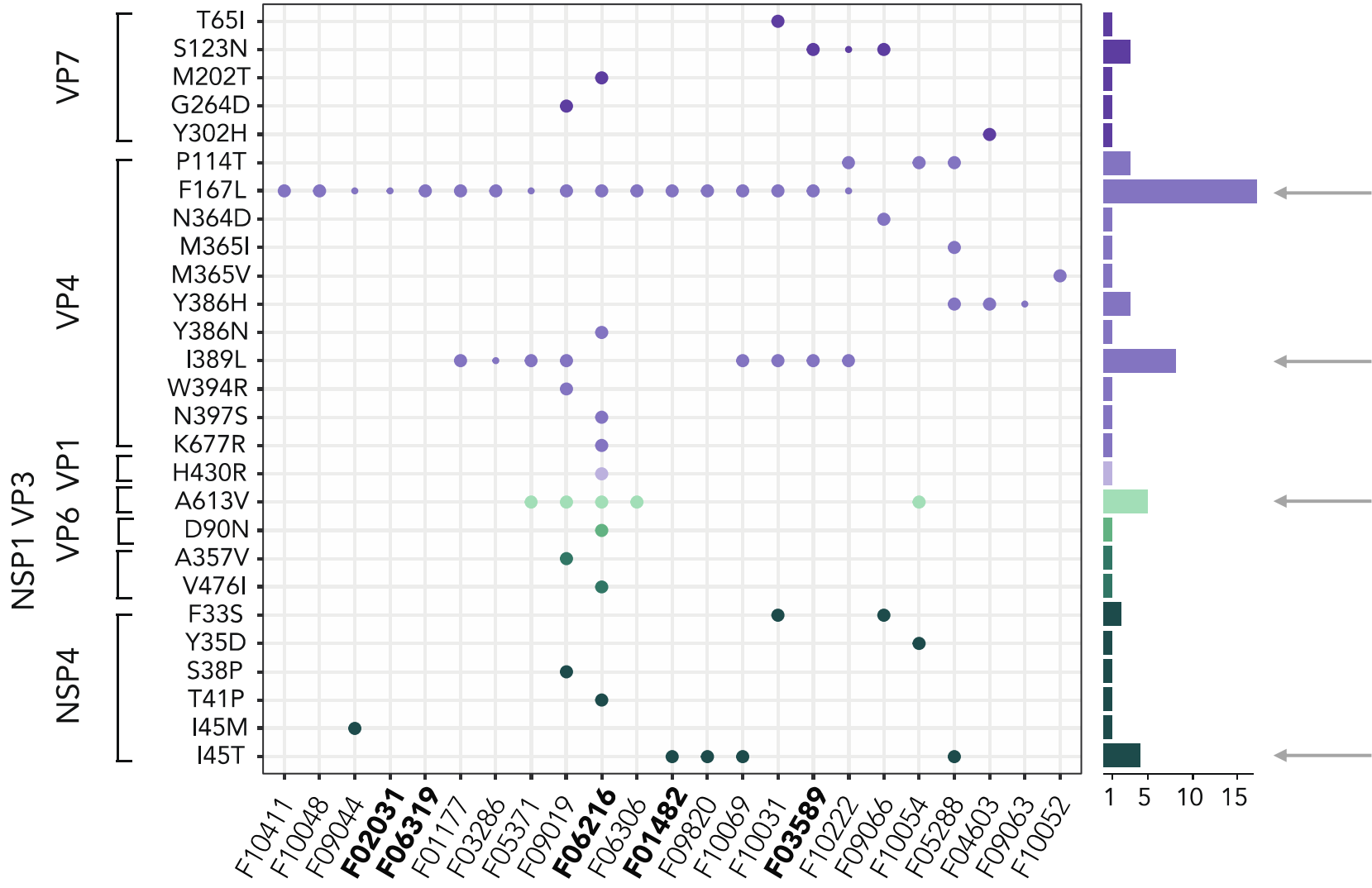


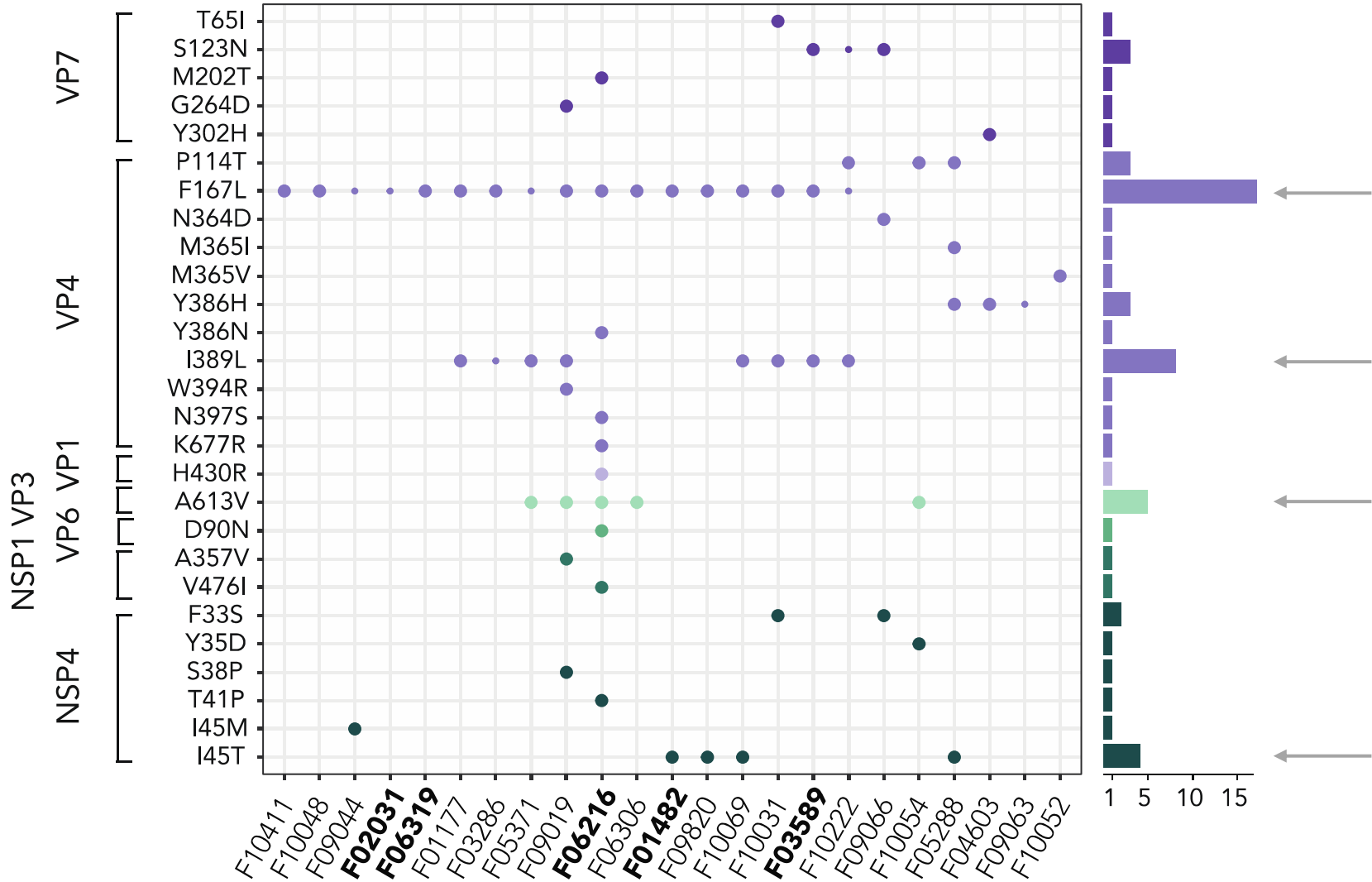




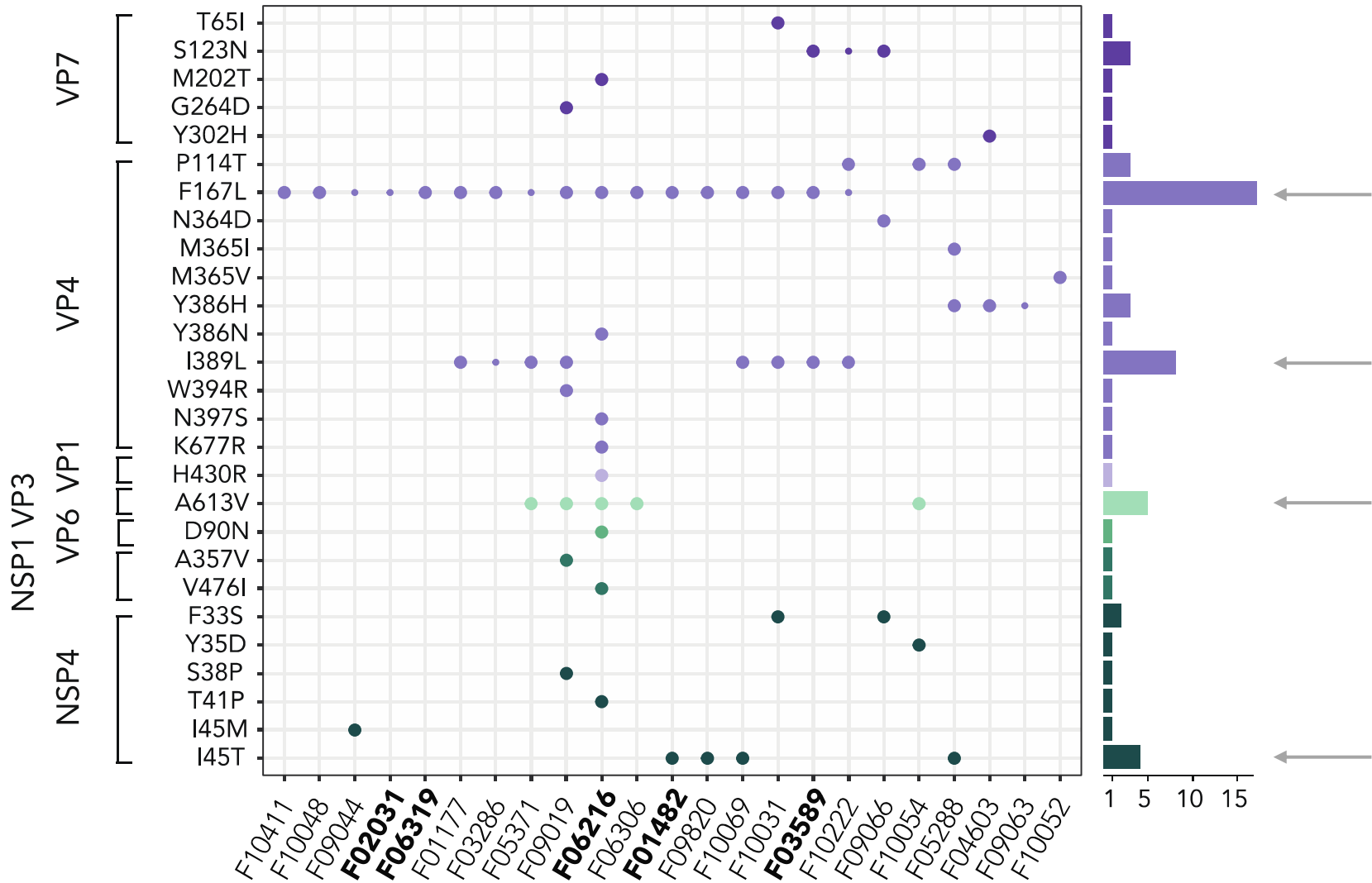
- VP4 harbors >50% mutations
- 88% of the point mutations were non-synonymous and accumulate rapidly







○ Reversion of vaccine strain?



- Reversion of vaccine strain?
- According to literature F167L and I45T could be reversions!

# Summary

- No evidence of reassortment of vaccine strain with wild type strains
- Approximately 40% of cases might be explained by other pathogens
- Very limited evidence for vaccine circulation in the human population
- Vaccine strains might revert rapidly to parental strains

# NRC Rotavirus

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Mandy Bloemen

Lize Cuypers