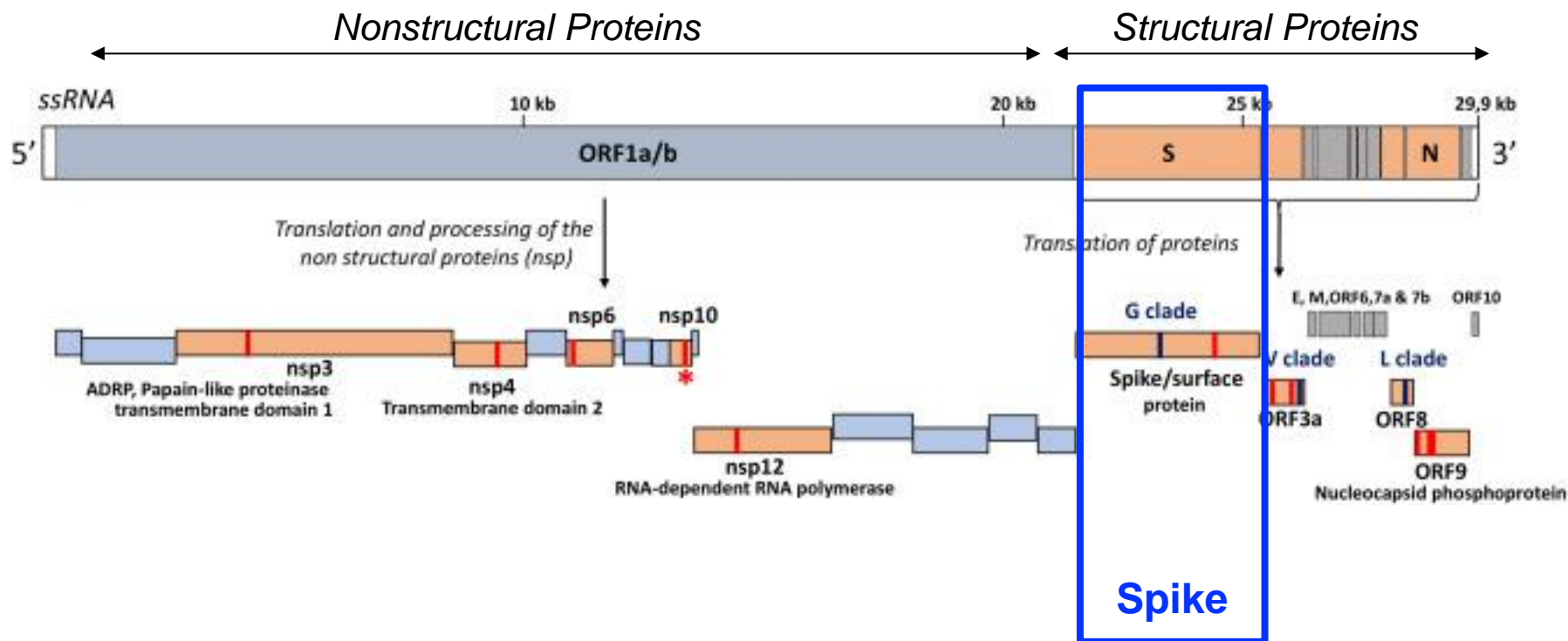


SARS-CoV-2 Genomdiversität

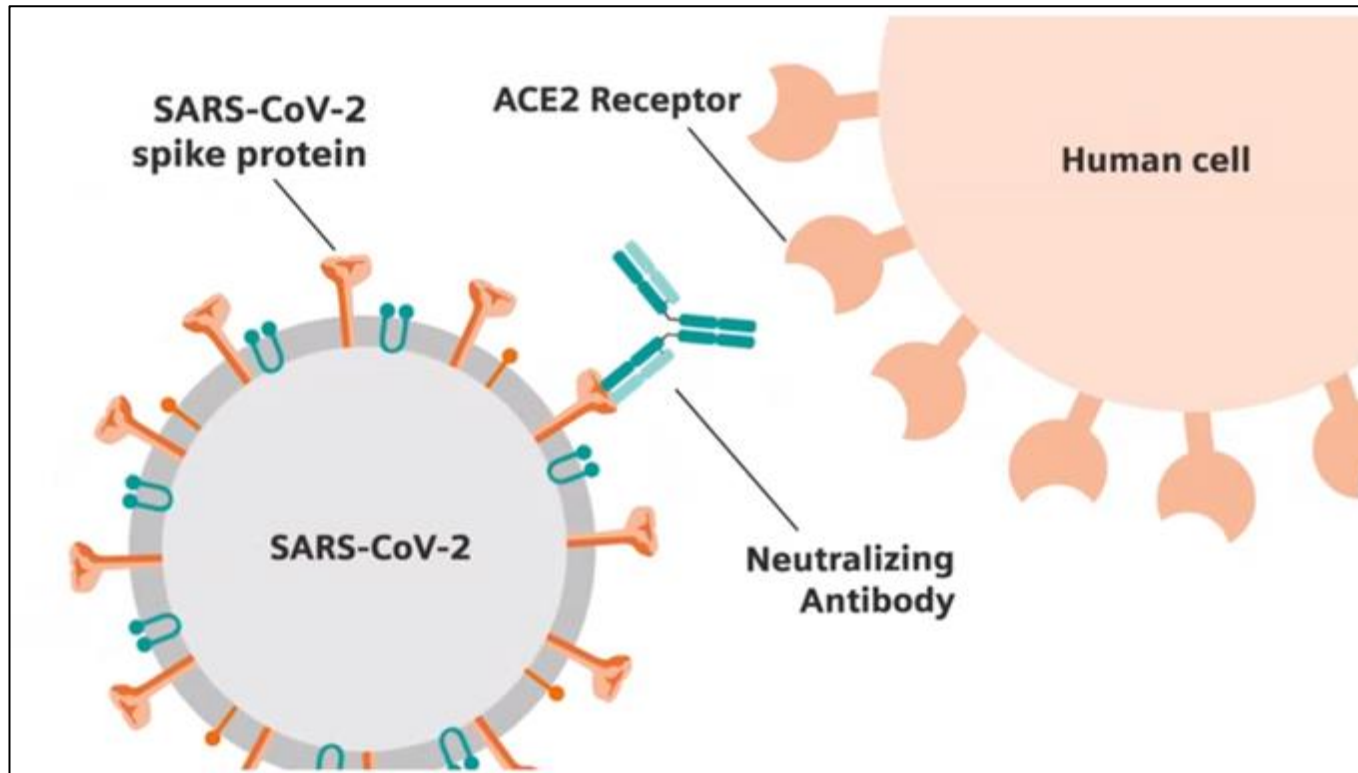
Update für den RKI Krisenstab, 3. August 2020

SARS-CoV-2 Genome



- (+)ssRNA Virus, ~30 kb
- SARS-CoV-2's RNA-abhängige Polymerase hat rudimentäre Korrekturlesefähigkeit → langsame Evolutionsrate → *Good News!*

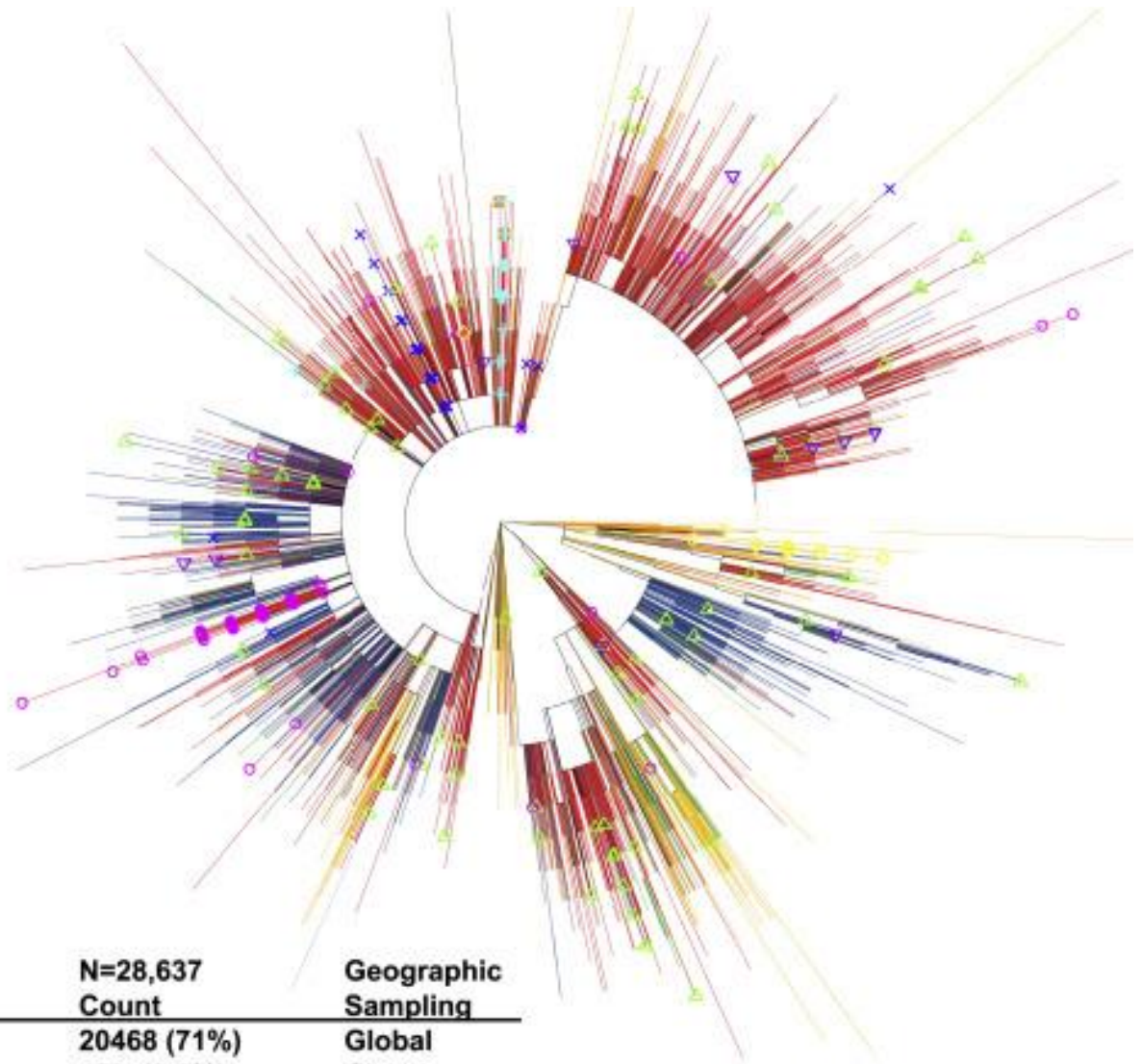
Rationale für Molekulare Surveillance des *Spike* Gens



1. Bindung an ACE-2 Rezeptor → Eintritt in die Zielzelle / Virulenz
2. Einfaches (!) Target für neutralisierende Antikörper („Achillesferse“) → Impfung & monoklonale Antikörper / Escape

Spike Molekulare Surveillance

A

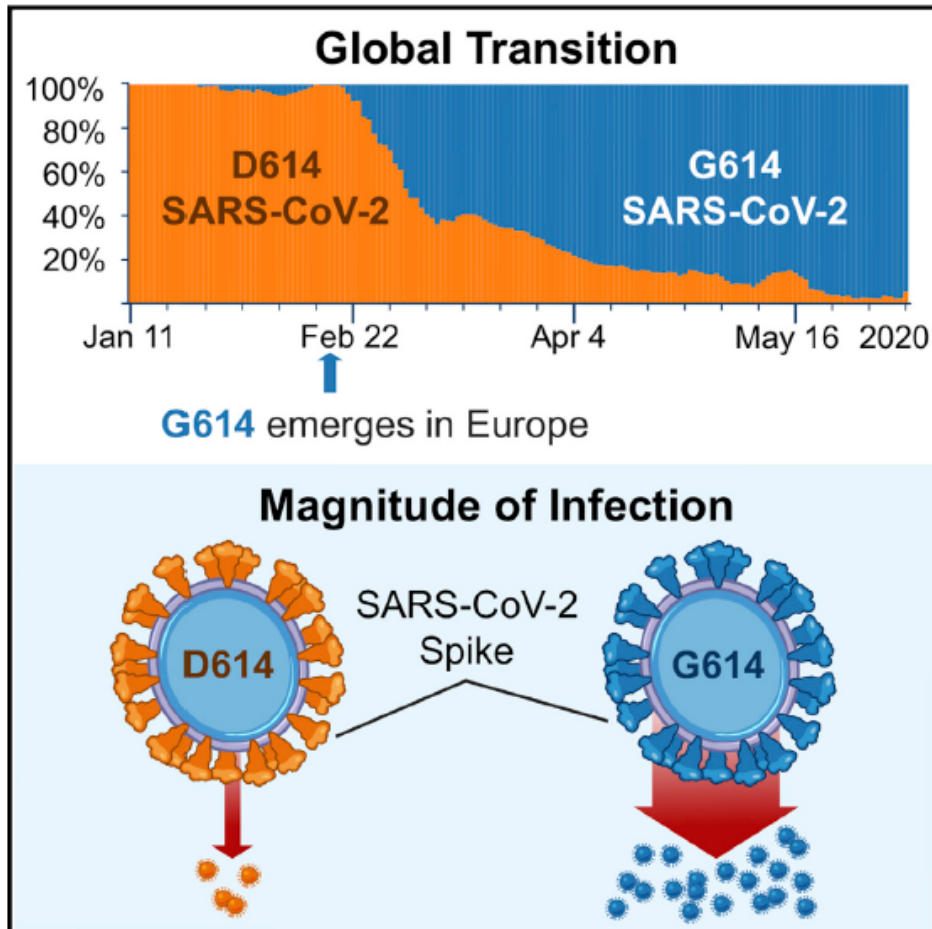


B

Spike Mutation	Spike Region Possible Impact	N=28,637 Count	Geographic Sampling
D614G	SARS-CoV epitope	20468 (71%)	Global
L5F	Signal Peptide	170 (0.6%)	Global
R21I/K/T	S1 NTD domain	133 (0.5%)	Wales, England, others
A829T/S	Fusion Peptide	91 (0.3%)	Thailand
D839Y/N/E	Fusion Peptide	149 (0.5%)	Portugal, New Zealand, others
D936Y/H	Heptad Repeat 1(HR1)	257 (0.9%)	Sweden, UK, others
P1263L	Cytoplasmic Tail	201 (0.7%)	UK, others

Tracking Changes in SARS-CoV-2 Spike: Evidence that D614G Increases Infectivity of the COVID-19 Virus

Bette Korber,^{1,2,10,*} Will M. Fischer,¹ Sandrasegaram Gnanakaran,¹ Hyejin Yoon,¹ James Theiler,¹ Werner Abfalterer,¹

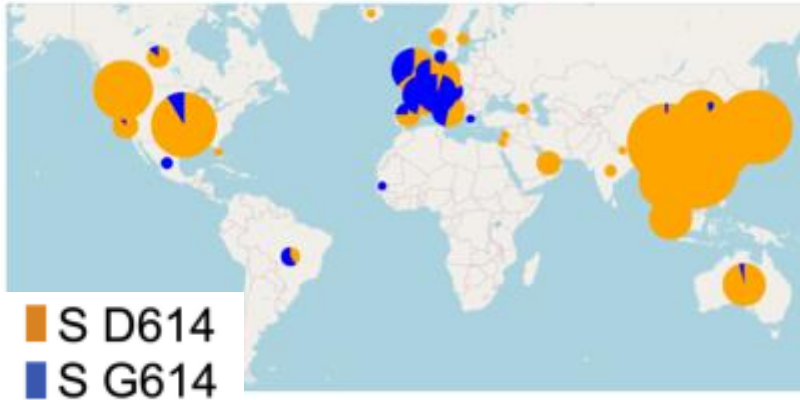


Highlights

- A SARS-CoV-2 variant with Spike G614 has replaced D614 as the dominant pandemic form
- The consistent increase of G614 at regional levels may indicate a fitness advantage
- G614 is associated with lower RT PCR Cts, suggestive of higher viral loads in patients
- The G614 variant grows to higher titers as pseudotyped virions

G614 ist zur weltweit vorherrschenden Variante geworden.

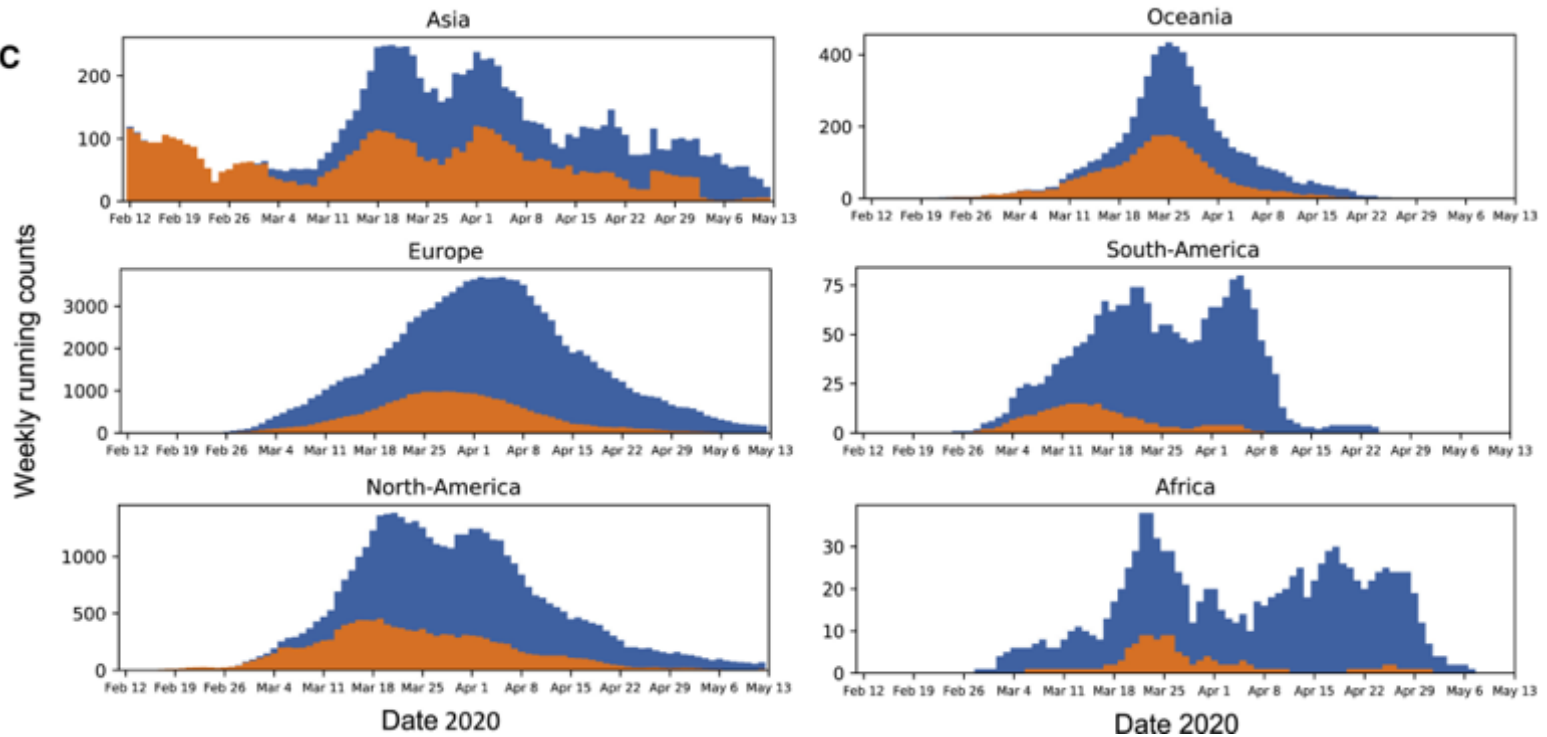
Prior to March 1



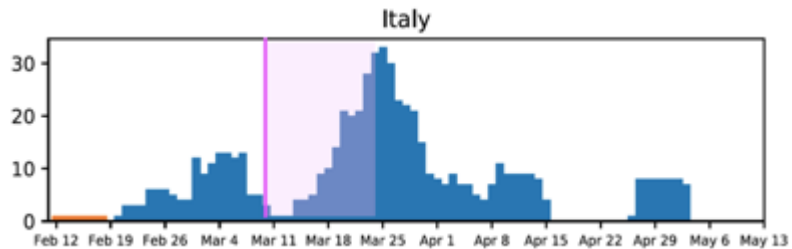
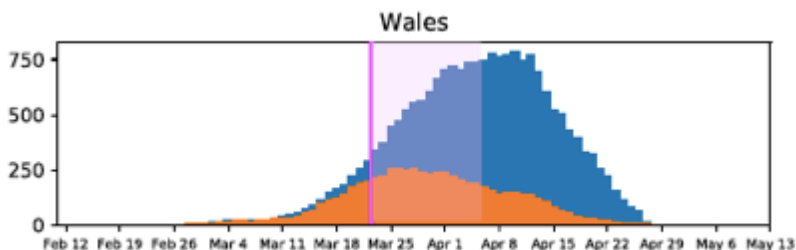
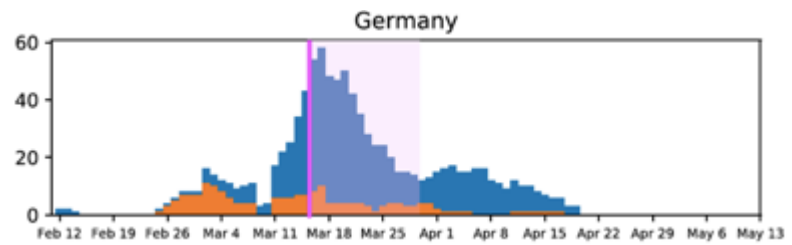
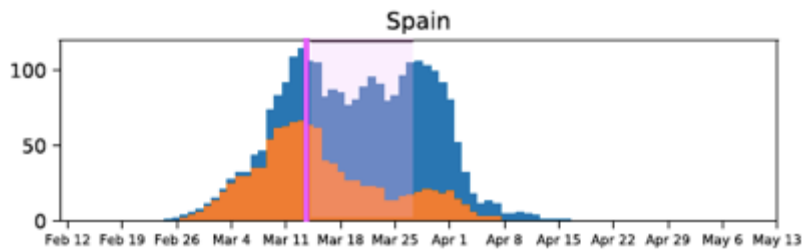
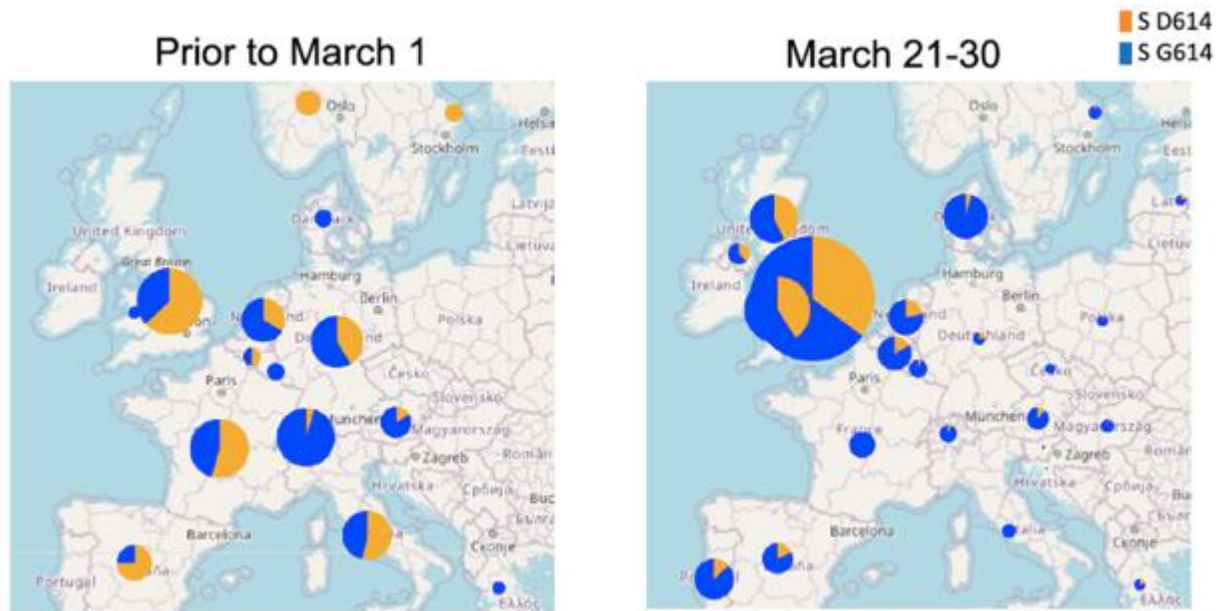
March 21-30



C



G614 ist in fast allen infizierten Populationen rasch dominant geworden, auch dort, wo initial D vorherrschte.



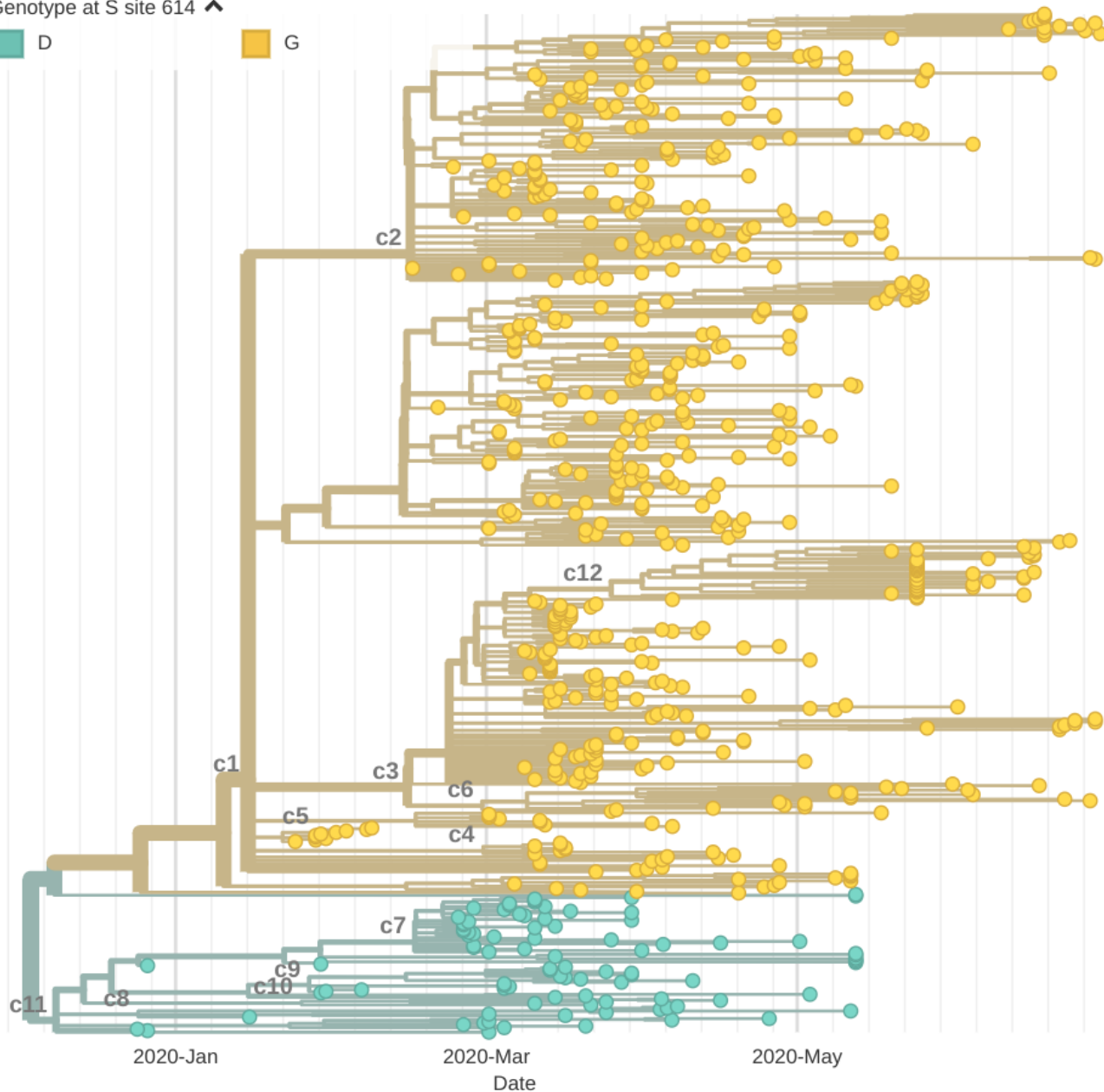
Lockdown= pink line, followed by a light pink block indicating max. 2-week incubation.

G614 kommt in 574 von 657 deutschen Genomen vor

Genotype at S site 614 ^

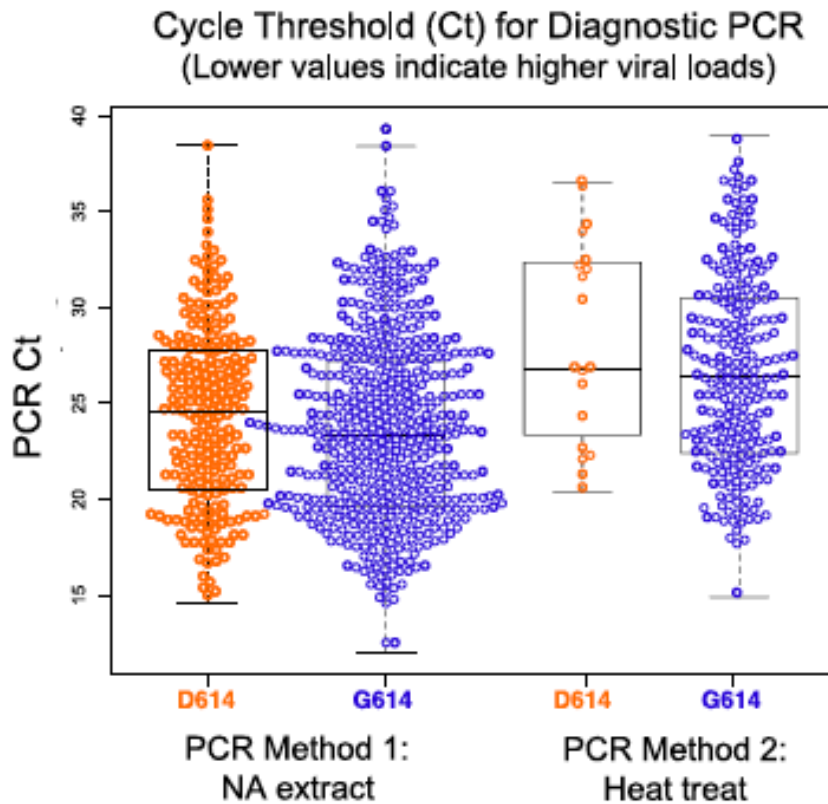
D

G

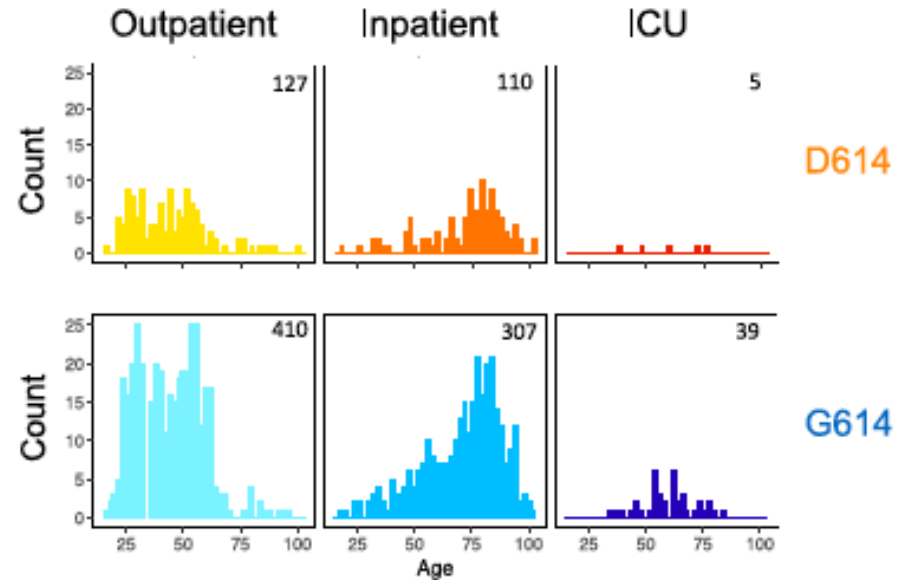


G614 ist mit höherer Viruslast assoziiert, aber nicht mit schwererer Erkrankung

A Ct D614 vs G614
GLM $p = 0.037$



B Hospitalization D614 vs G614
Fisher' $p = 0.66$



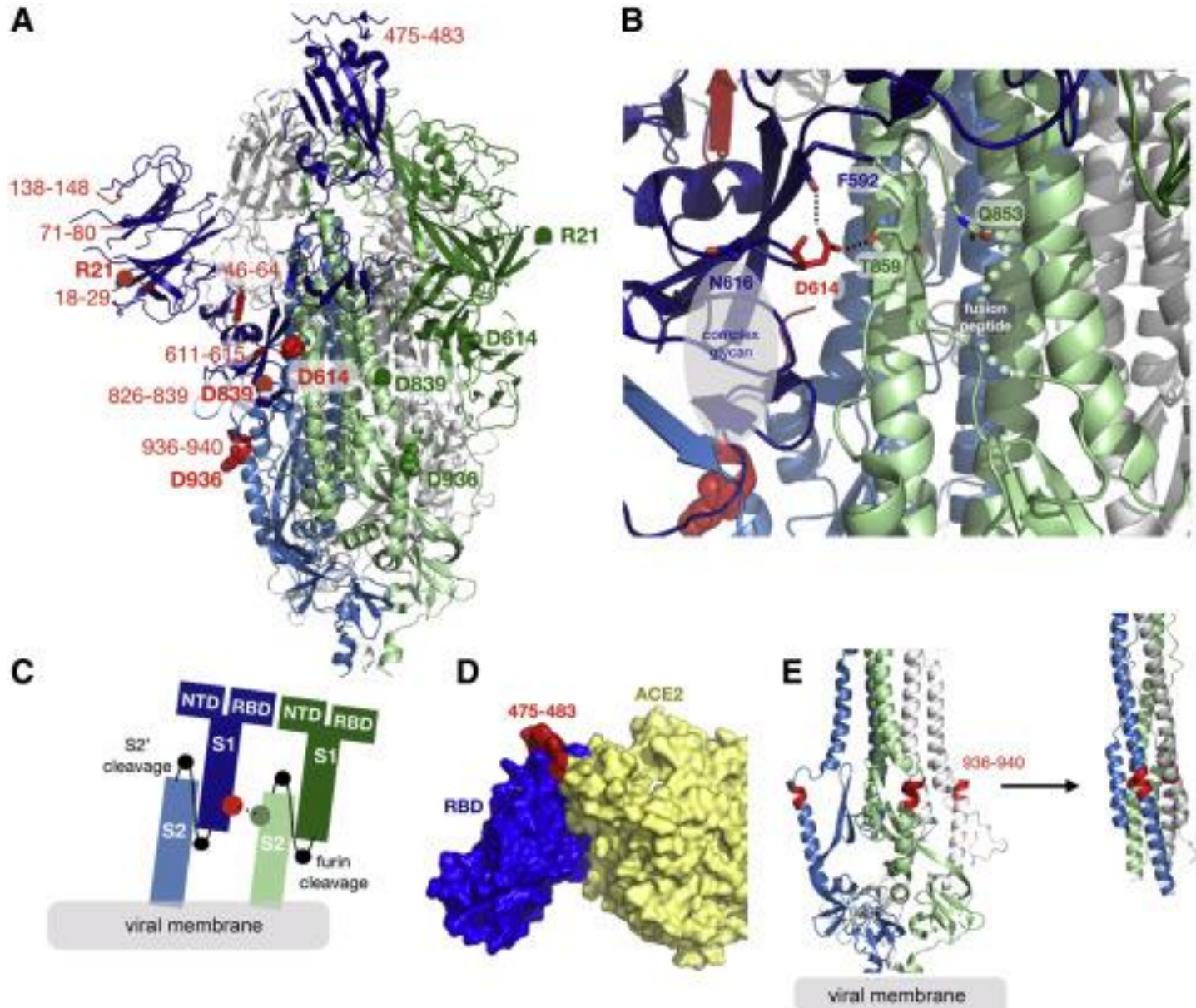
	OP	IP	ICU
D614	127	110	5
G614	410	307	39

Fisher's exact, 2x2: (OP+IP) x ICU = 0,047

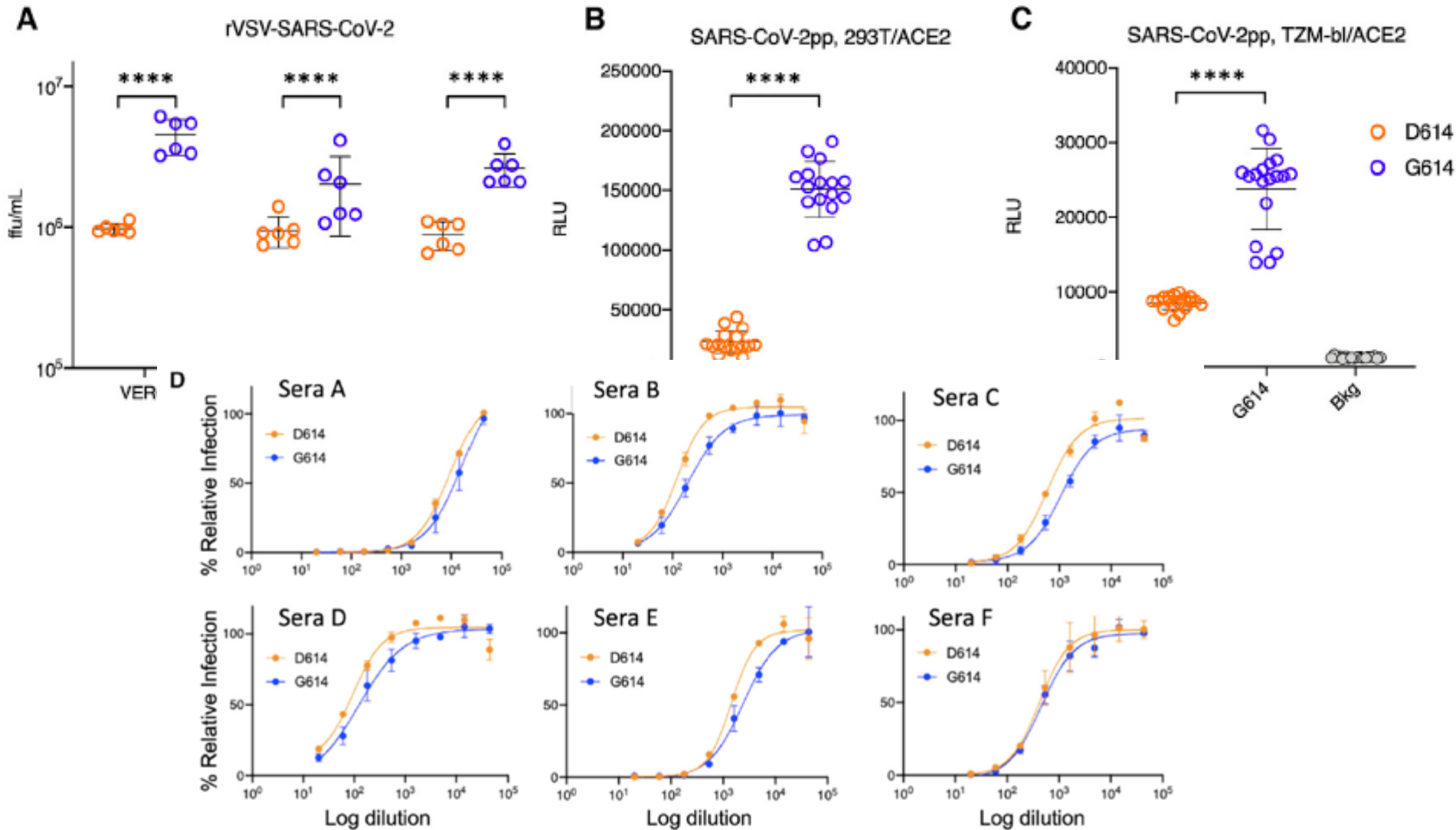
Fisher's exact, 2x2, OP x (IP+ICU): $p = 0.66$

Diese Daten wurden von einer anderen Gruppe bestätigt (T. Bedford, 2020)

G614 könnte den Spike stabilisieren

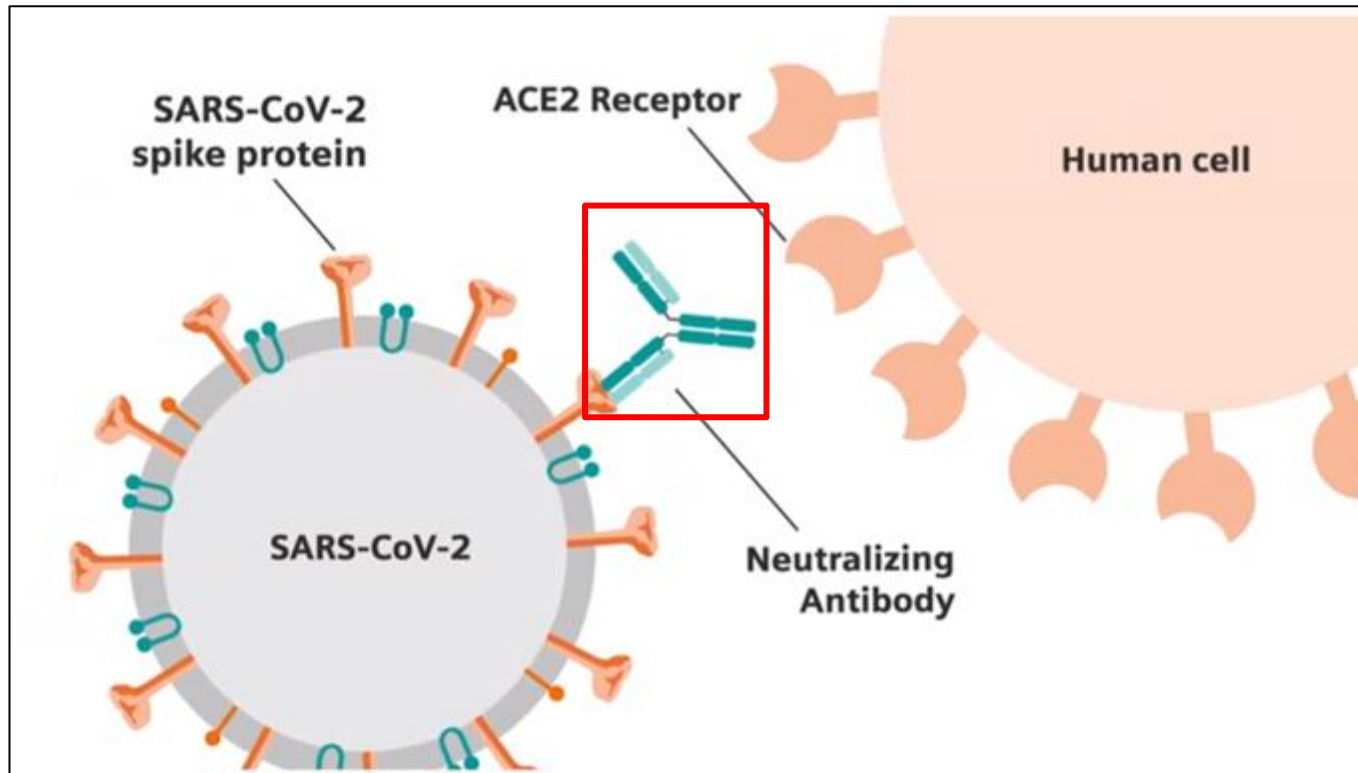


Mögliche funktionale Relevanz von G614: Infektiöser (Pseudoviren), kein Einfluss auf Neutralisierung



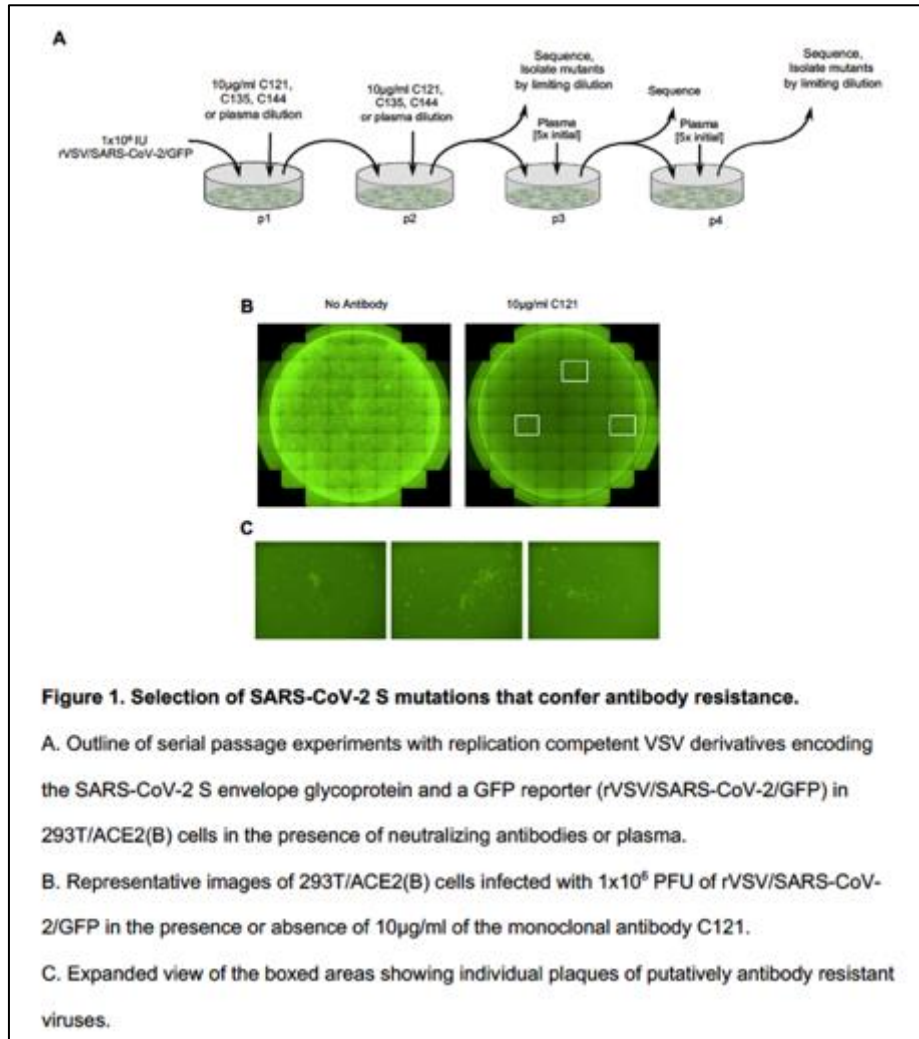
Diese Daten wurden von einer anderen Gruppe bestätigt (Zhang, 2020)

Rationale für Molekulare Surveillance des *Spike* Gens



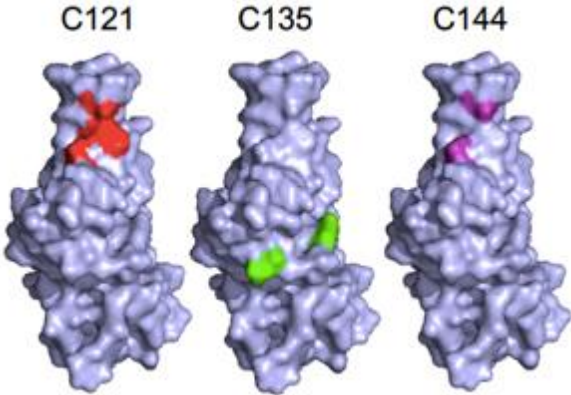
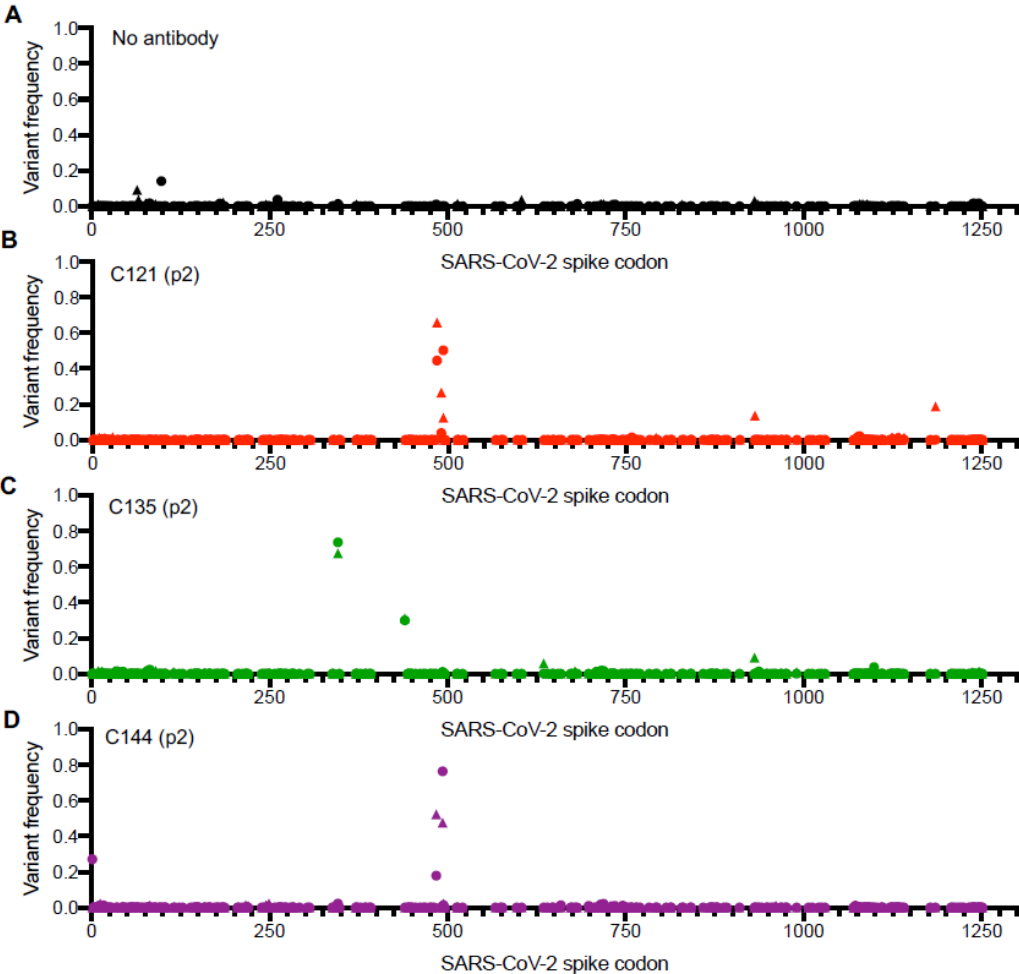
1. Bindung an ACE-2 Rezeptor → Eintritt in die Zielzelle / Virulenz
2. Einfaches (!) **Target für neutralisierende Antikörper** („Achillesferse“) → Impfung & monoklonale Antikörper / Escape

Spike Variation: Ist ein Escape vor neutralisierenden Antikörpern möglich?

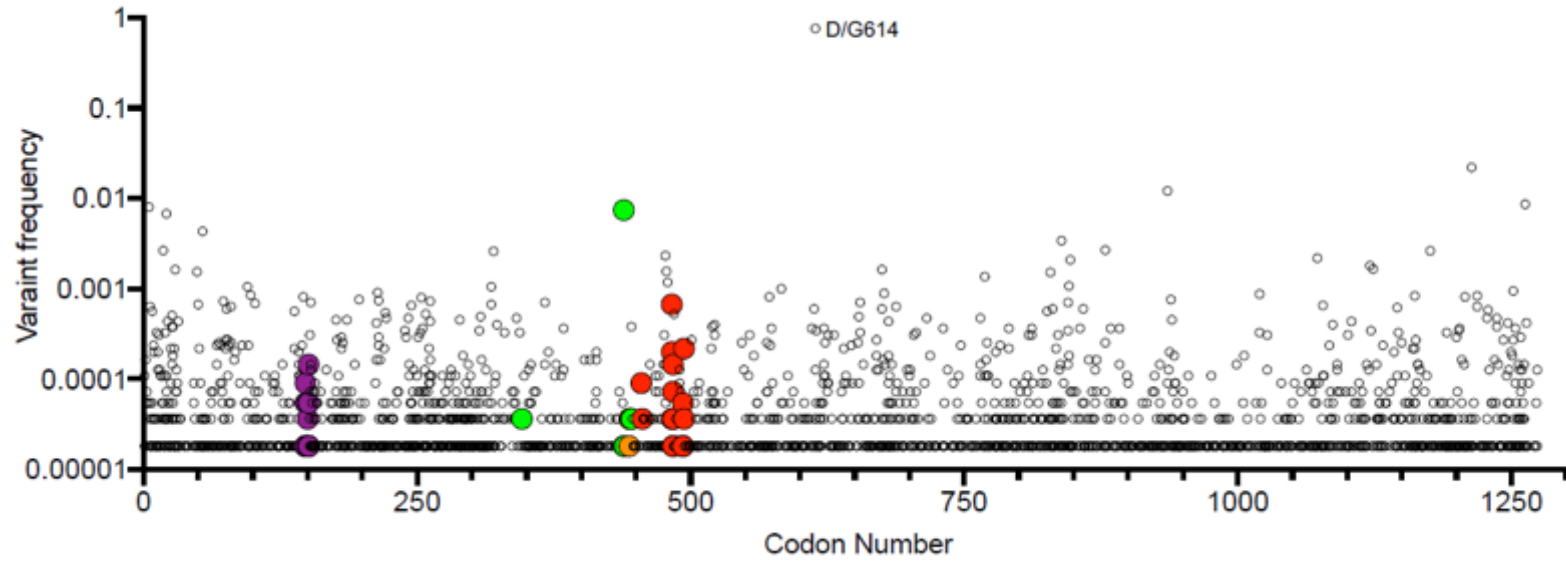


Selektion von Spike-Mutanten *in vitro* durch Anzucht von (chimerischen rVSV/SARS-CoV-2) Viren in Gegenwart neutralisierender Antikörper bzw. rekonvaleszenten Plasmas

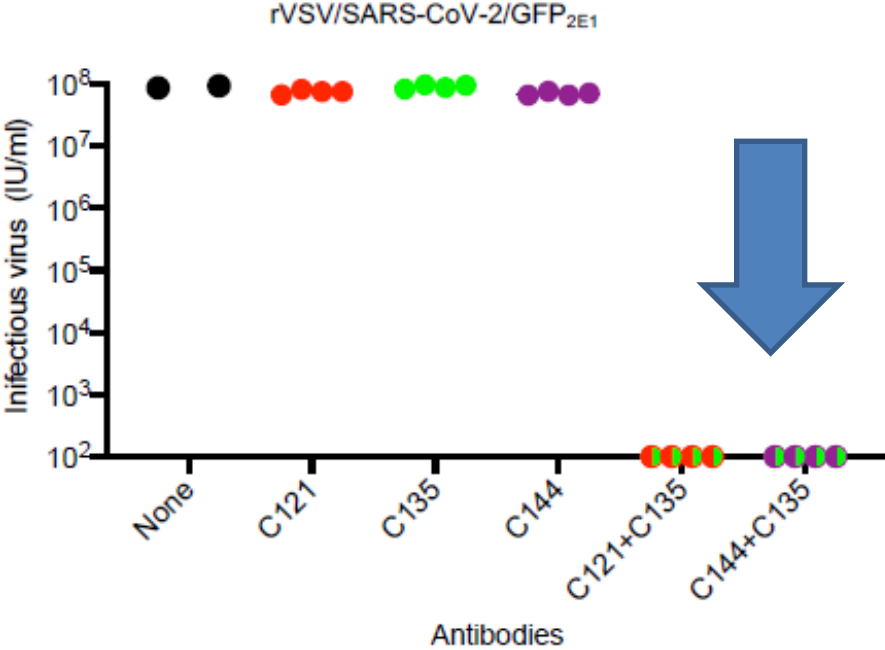
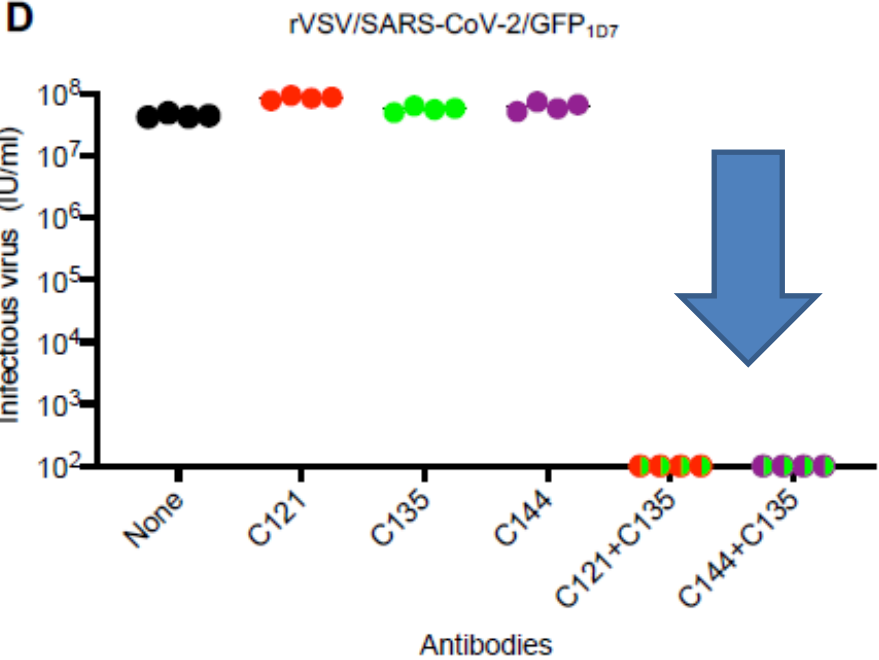
Selektion von Spike-Mutanten in Gegenwart neutralisierender Antikörper



Resistente Spike-Mutanten kommen in niedriger Frequenz in der Natur vor



Antikörper-Resistenz lässt sich durch Einsatz von AK-Kombinationen vollständig überwinden.

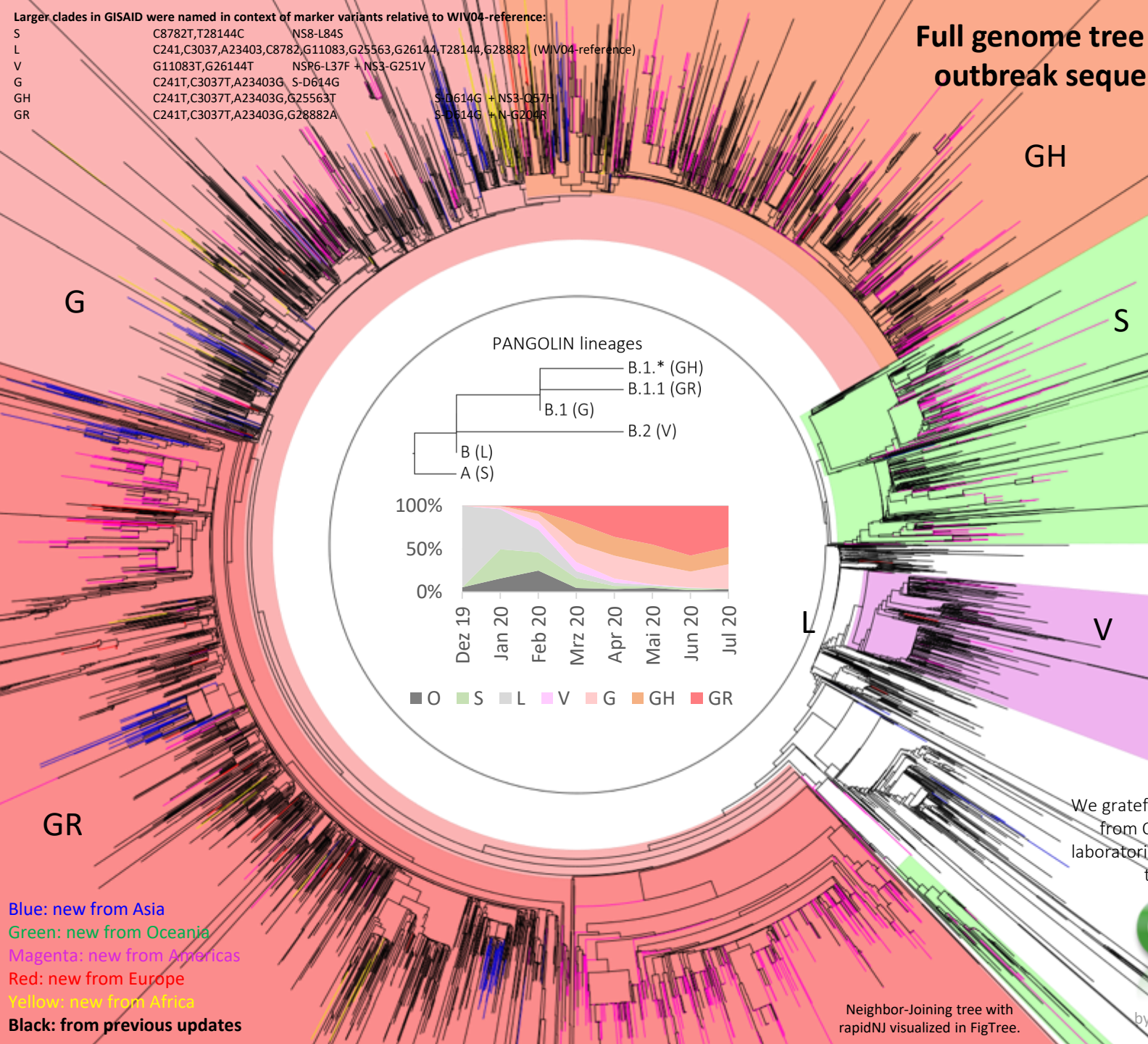


SARS-CoV-2 Clades

Larger clades in GISAID were named in context of marker variants relative to WIV04-reference:

S	C8782T,T28144C	NS8-L84S
L	C241,C3037,A23403,C8782,G11083,G25563,G26144,T28144,G28882	(WIV04-reference)
V	G11083T,G26144T	NSR6-L37F + NS3-G251V
G	C241T,C3037T,A23403G	S-D614G
GH	C241T,C3037T,A23403G,G25563T	S-D614G + NS3-Q57H
GR	C241T,C3037T,A23403G,G28882A	S-D614G + N-G204R

Full genome tree derived from all outbreak sequences 2020-07-29



Notable changes:
67,103 full genomes (+2,471) (excluding low coverage, out of 72,942 entries)

Updated clades:

- S clade 4,574 (+279)
- L clade 3,707 (+4)
- V clade 4,252 (+11)
- G clade 15,611 (+293)
- GR clade 21,187 (+1314)
- GH clade 14,689 (+486)
- Other clade 3,083 (+84)

We gratefully acknowledge the Authors from Originating and Submitting laboratories of sequence data on which the analysis is based.



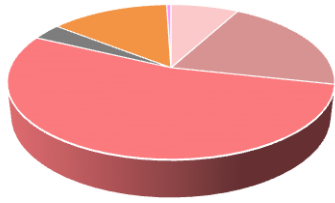
by BII/GIS, A*STAR Singapore

Blue: new from Asia
 Green: new from Oceania
 Magenta: new from Americas
 Red: new from Europe
 Yellow: new from Africa
 Black: from previous updates

Neighbor-Joining tree with rapidNJ visualized in FigTree.

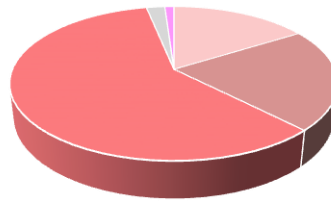
Regional clade distribution of new sequences 2020-07-29

North America (+1991)



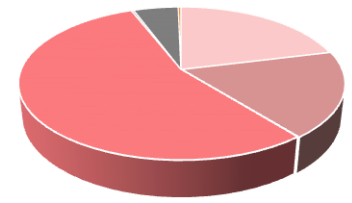
■ G ■ GH ■ GR ■ L ■ O ■ S ■ V

Europe (+93)



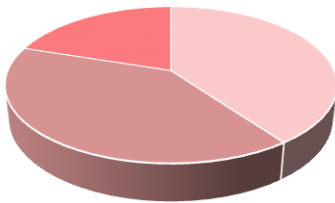
■ G ■ GH ■ GR ■ L ■ O ■ S ■ V

Asia (+303)



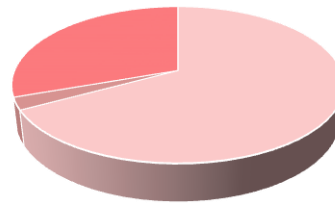
■ G ■ GH ■ GR ■ L ■ O ■ S ■ V

South America (+5)



■ G ■ GH ■ GR ■ L ■ O ■ S ■ V

Africa (+79)



■ G ■ GH ■ GR ■ L ■ O ■ S ■ V

Oceania (+0)

■ G ■ GH ■ GR ■ L ■ O ■ S ■ V

S clade 4,574 (+279): 128 Panama, 126 USA/WA, 19 USA/MN, 4 USA/FL, 1 India, 1 USA/MI

L clade 3,707 (+4): 2 England, 1 USA/MN, 1 Lishui

V clade 4,252 (+11): 4 USA/MN, 3 USA/FL, 2 Panama, 1 England, 1 USA/VA

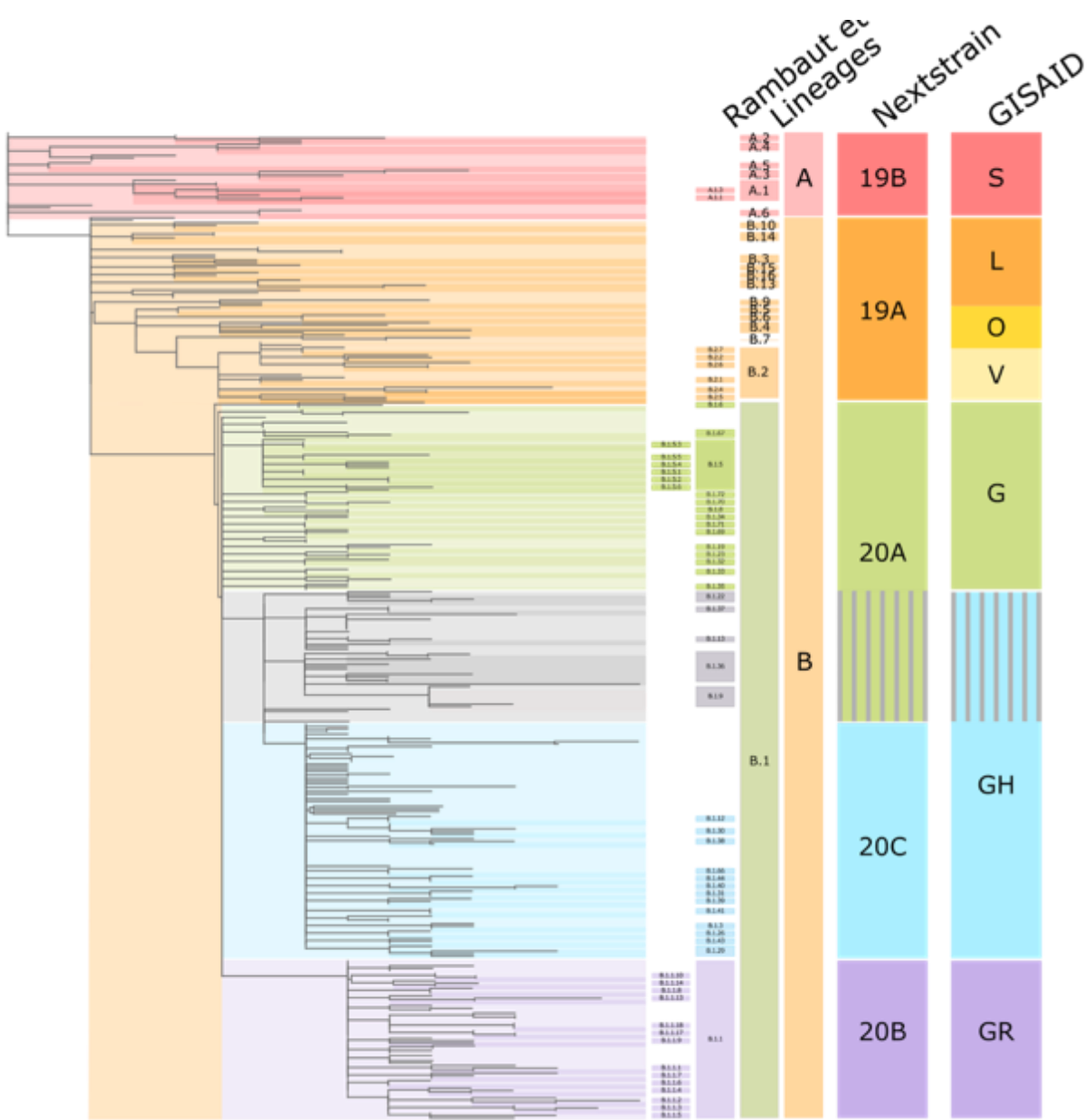
G clade 15,611 (+293): 64 India, 58 USA/WA, 36 DRC, 31 Mexico, 31 Panama, 26 USA/WI, 17 South Africa, 6 Italy, 6 England, 3 USA/MN, 3 Turkey, 3 USA/FL, 3 USA/CA, 2 Colombia, 1 USA/NJ, 1 Canada, 1 USA/IL, 1 USA/MI

GR clade 21,187 (+1314): 1037 USA/WA, 166 India, 28 Turkey, 26 England, 22 South Africa, 13 USA/CA, 6 USA/WI, 4 USA/MN, 3 USA/FL, 3 Panama, 2 DRC, 1 Italy, 1 USA/VA, 1 Mexico, 1 Brazil

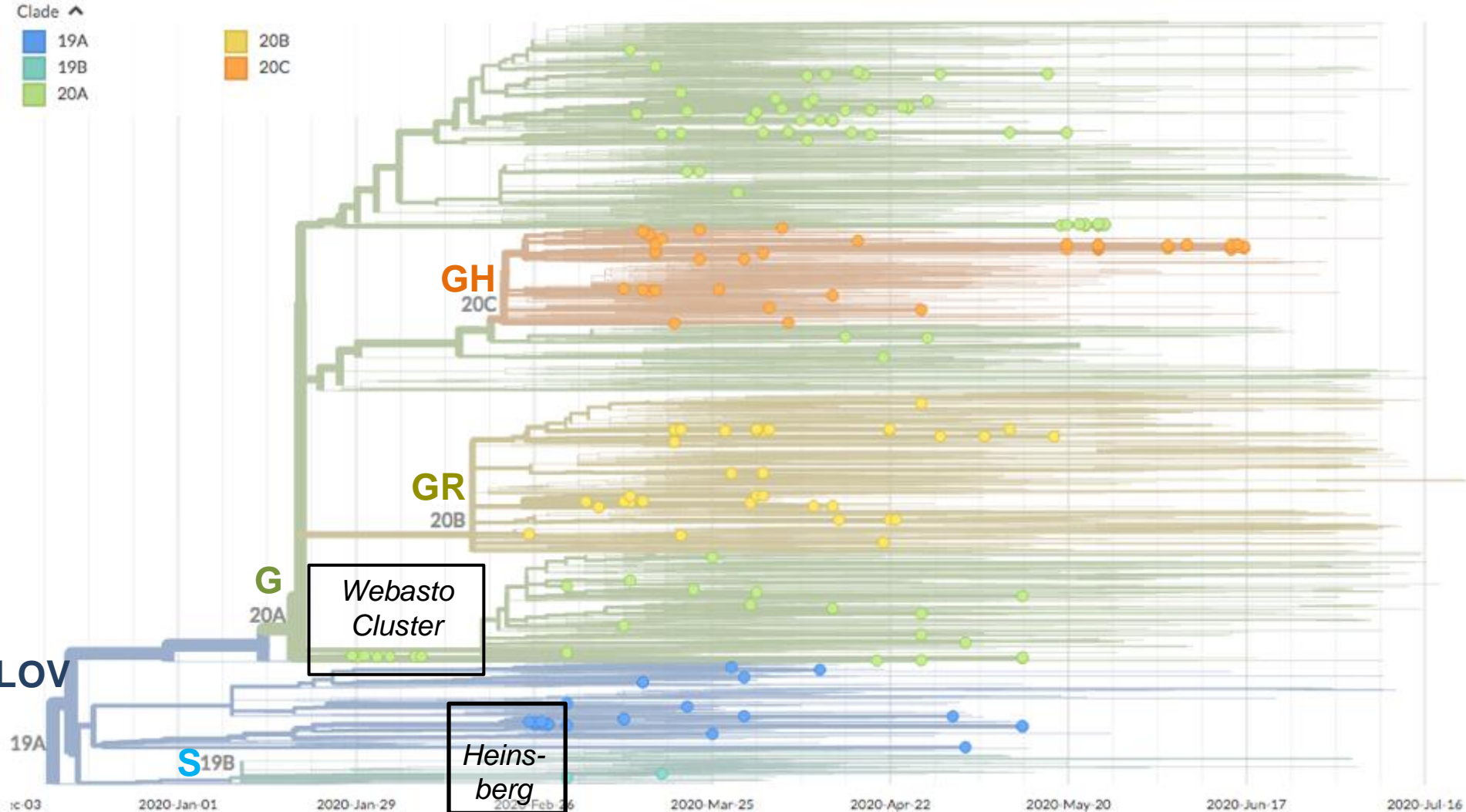
GH clade 14,689 (+486): 228 USA/WA, 55 India, 45 USA/FL, 32 USA/MI, 30 USA/VA, 24 USA/CA, 16 USA/MN, 16 Turkey, 14 USA/WI, 6 Mexico, 6 Panama, 4 England, 2 DRC, 2 Canada, 2 Colombia, 1 USA/SC, 1 USA/MO, 1 USA/IL, 1 USA/NM

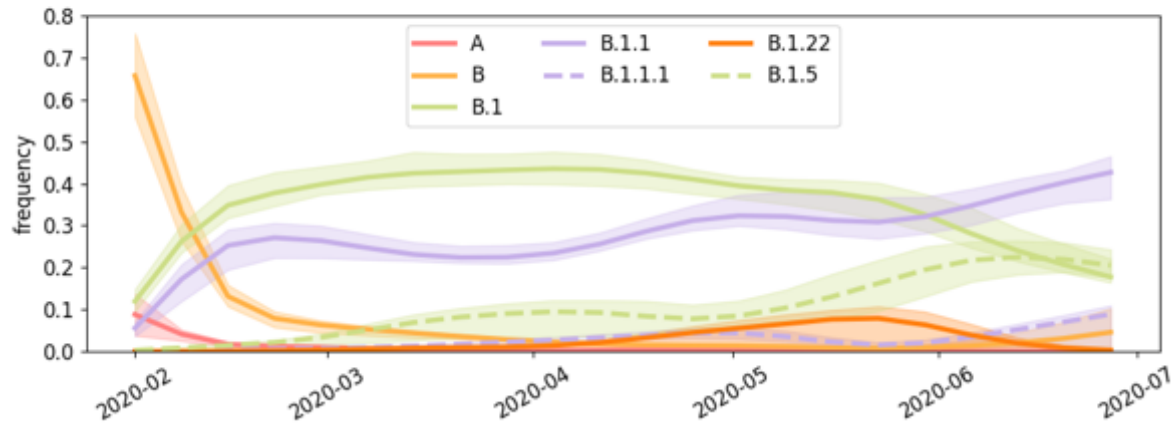
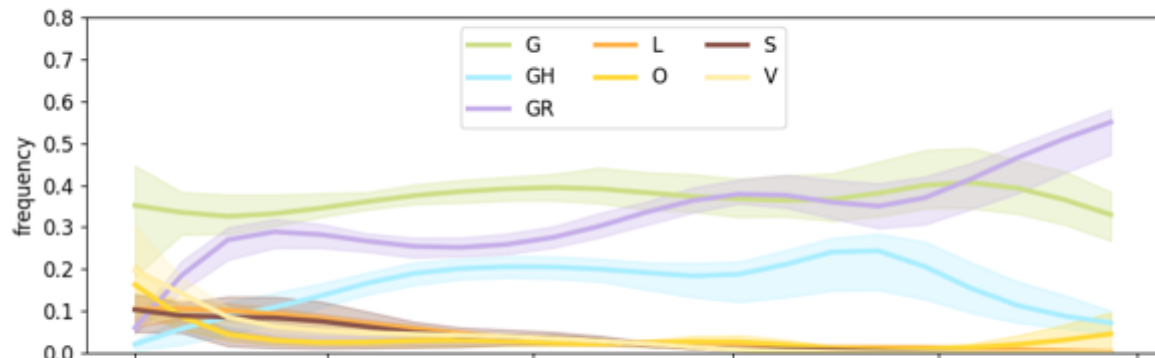
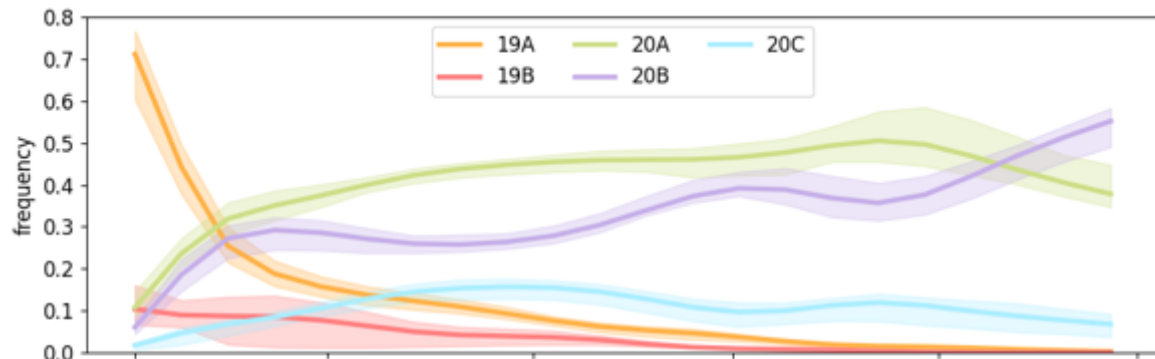
O clade 3,083 (+84): 56 Panama, 16 India, 11 USA/WA, 1 USA/FL

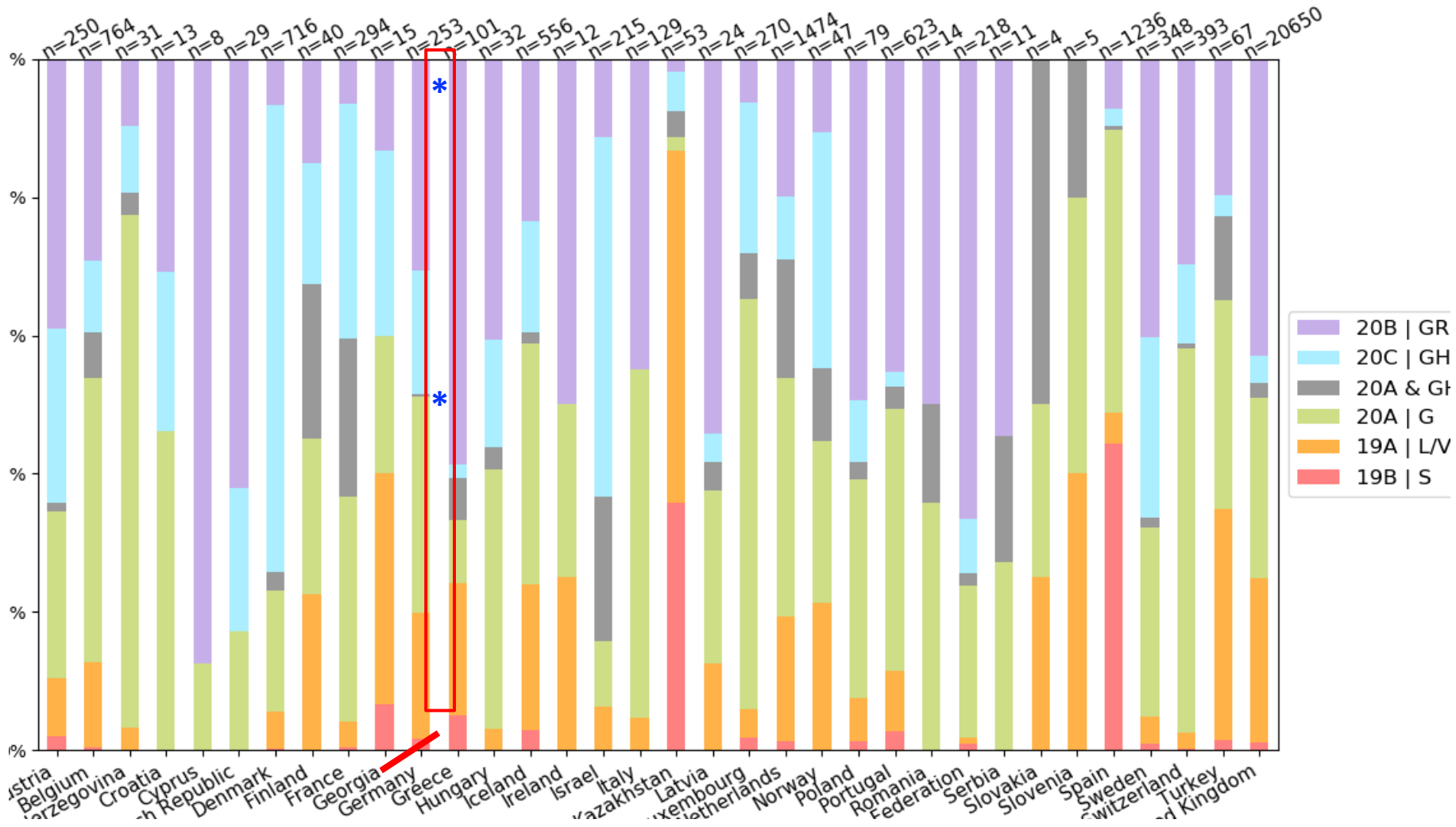
Region	Total	New	Region	Total	New	Region	Total	New
Europe	41,193	93	Africa	979	79	North America	16,053	1,991
Asia	5,552	303	Oceania	2,490	0	South America	836	5



German Sequences: 19A, 19B, 20A, 20B, 20C







* RKI Sequences on Nextstrain belong to Clades G/20A and GR/20B