Neutralizing Activity of Sera from Sputnik V Vaccinated People against Variants of Concern (VOC: B.1.1.7, B.1.351, P.1, B.1.617.2, B.1.617.3) and Moscow Endemic SARS-CoV-2 Variants

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Supplementary

Supplementary methods
Nasopharyngeal swabs collection

The enrollment of patients with a verified diagnosis of COVID-19 and the collection of biological material during the disease progression was conducted at the Moscow City Infectious Disease Clinical Hospital No.1 Moscow Healthcare Department from May to June 2020 (the first wave of the pandemic) and from November 2020 to March 2021 (the second wave of the pandemic). Biomaterial was collected and transported in accordance with the requirements of Sanitary Norms and Regulations of the Russian Federation №1.3.3118-13 and №1.2.036-95. The patients under observation signed informed consent for biological material sampling and underwent survey to gather data for their subsequent use for scientific purposes. The study was approved by the ethics committee (minutes of the Local Ethics Committee No. 2a of May 11, 2020, No. 11a of November 16, 2020, and No. 1 of February 11, 2021).

The material collection was carried out from 1,572 patients (500 subjects during the first wave of the pandemic and 1,072 subjects during the second wave) diagnosed with COVID-19 who were hospitalized at different times from the onset of their first symptoms. The criterion for swab collection was a positive PCR result at the admission to the hospital. Biomaterial (nasopharyngeal swab) was collected from the patients examined by quantitative reverse transcription PCR. SARS-CoV-2 RNA was identified in the samples using a reagent kit for extraction and qualitative determination of SARS-CoV-2 coronavirus RNA using the SARS-CoV-2 FRT RT-PCR method, manufactured by Gamaleya National Research Institute of Epidemiology and Microbiology.

Spike-pseudotyped lentivirus

We have developed a lentivirus-based pseudovirus carrying the GFP reporter gene and S protein of SARS-CoV-2 on the surface of pseudovirus particles. Variants of pseudovirus, carrying the S proteins of the reference Wuhan strain and VOC B.1.1.7 and B.1.351 lineages variants were obtained. In order to evaluate the VNA on a pseudovirus, we have formed a serum panel of volunteers who previously received Sputnik V (table S4 and S5). The serum panel of 8 volunteers receiving two doses of Sputnik V vaccine (table S4) has been tested for neutralizing activity against pseudo-SARS-CoV-2 Wuhan, B.1.1.7 and B.1.351 lineages

Figure S1. **Prevalence of RBD mutations in Russian SARS-CoV-2 variants collected since 2021.** Colored bars represent frequencies of RBD mutations found in Russian SARS-CoV-2 genomes available in GISAID (blue) and the current study (orange) with sample collection dates starting from 2021.
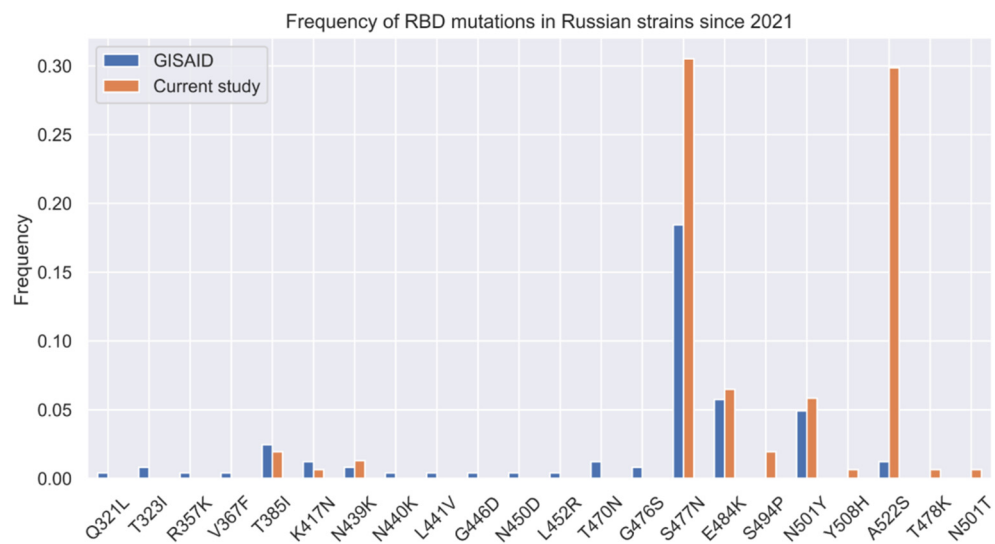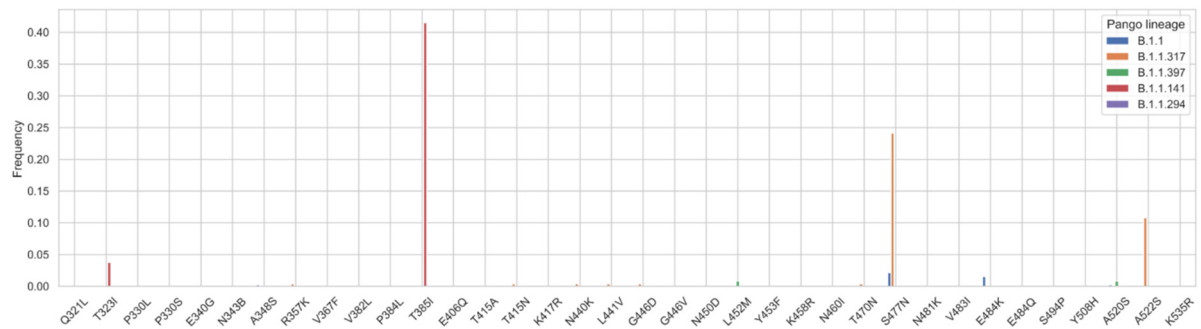
***Figure S2.* RBD mutations profiles of the major Russian SARS-CoV-2 genetic lineages**

Colored bars represent RBD mutation frequencies in Russian GISAID sequences for 5 genetic lineages majorly spread in Russia.

The estimation of variability according to our data in Moscow study shows that mutations S477N and A522S are coupled in 98% of cases (Pearson correlation coefficient 0.99) (figure S3). In contrast, genetic data for Russian variants sequences from GISAID shows that S477N mutation is mostly present separately from A522S.

Figure S3: **Correlation matrix of co-presence of mutations in the RBD**. Numbers in cells represent the Pearson coefficient for one-hot encoded mutations in RBD according to the data of the current study.

Figure S4: **Neutralization activity of serum samples after two doses of the Sputnik V vaccine recipients against wild-type pseudo-SARS-CoV-2 and B.1.1.7 or B.1.351 spike mutant viruses**. Dose-response curves of neutralizing activity of serum against (A) Wuhan reference strain, (B) lineage B.1.1.7 and (C) lineage B.1.351 SARS-CoV-2 pseudoviruses. Data represent mean value ± SEM of n=3 technical replicates. The numbers (003-013) indicate individual sera.
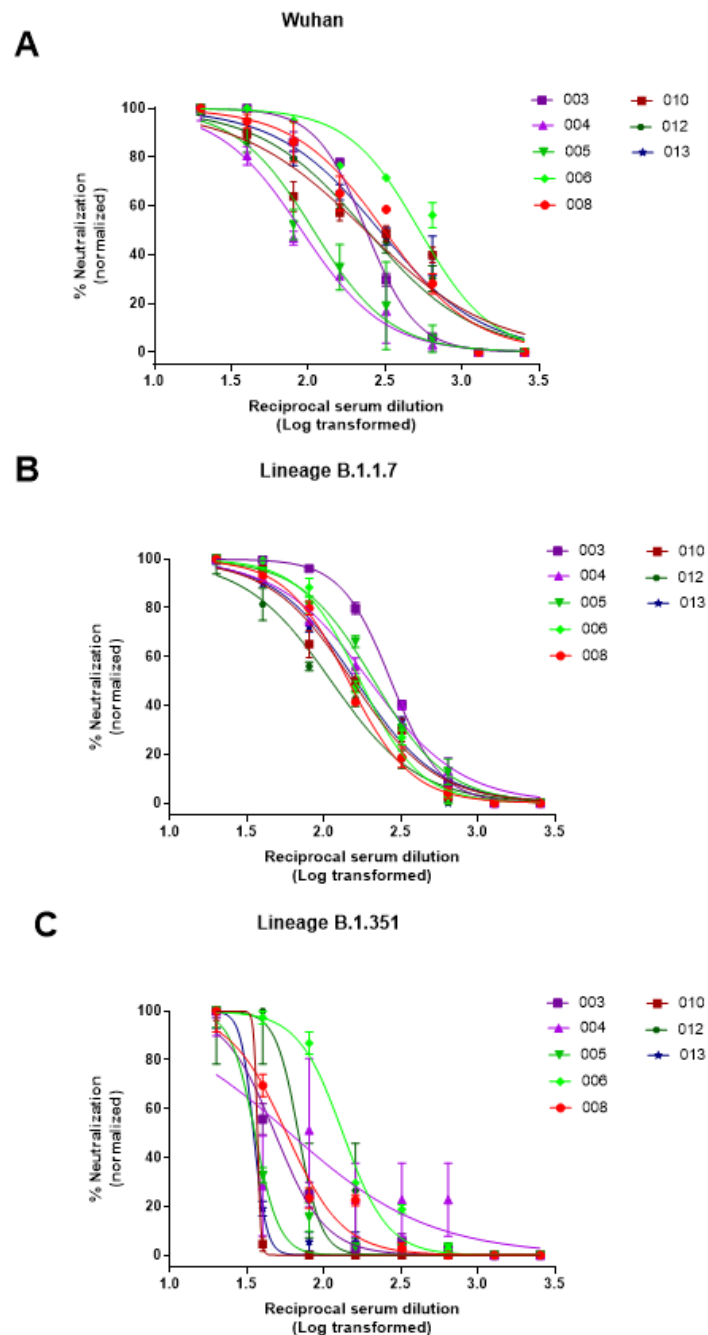
Table S1: The most common mutations in the RBD as reported in this study

Position in Spike	385	417	439	477	478	484	494	501	508	522	Amount of appearance in current research (Nov-Mar 2021)	
Wild type	T	K	N	S	T	E	S	N	Y	A	94	55,62%
S477N + A522S	·	·	·	N	·	·	·	·	·	S	47	27,81%
N501Y	·	·	·	·	·	·	·	Y	·	·	8	4,73%
E484K	·	·	·	·	·	K	·	·	·	·	6	3,55%
T385I	I	·	·	·	·	·	·	·	·	·	4	2,37%
E484K + S494P	·	·	·	·	·	K	P	·	·	·	3	1,77%
N439K	·	·	K	·	·	·	·	·	·	·	2	1,18%
K417N + E484K + N501Y	·	N	·	·	·	K	·	Y	·	·	1	0,59%
S477N	·	·	·	N	·	·	·	·	·	·	1	0,59%
T478K	·	·	·	·	K	·	·	·	·	·	1	0,59%
N501T	·	·	·	·	·	·	·	T	·	·	1	0,59%
Y508H	·	·	·	·	·	·	·	·	H	·	1	0,59%

Table S2: Putative phenotypic effect of dominating RBD mutations

Mtation	Increased binding affinity to hACE2 receptor	Escape from some mAbs	Escape some convalescent/vaccinated sera	Increased transmissi-bility	More severe illness	References
T385I	✓					1
K417N	✓	✓				1, 15
K417N+ +E484K+ +N501Y	✓	✓	✓	✓		8, 9, 10, 15
N439K	✓	✓	✓	✓		1, 4
S477N	✓ Slightly					1, 13
T478K	✓ In silico prediction	✓				11, 12
E484K	✓	✓	✓			1, 2, 9
S494P	✓	✓				16, 17
N501Y	✓	✓ Not critical	✓ Not critical	✓	✓	1, 3, 5, 6, 7, 10
N501T	✓					1
Y508H		✓				14
A522S	✓					1

Table S3: Characteristics of volunteers' sera used in the study of VNA in a pseudovirus model

Sera ID	Days from the 1st dose	Days from the 2nd dose	COI* $_{RBD}$	COI $_{Nc}$	Sex	Age
003	147	108	10.8	0.3	F	30-35
004	64	44	6.4	0.4	F	25-30
005	64	45	9.3	0.5	F	35-40
006	136	108	9.1	0.4	F	30-35
008	64	45	9.6	0.3	F	35-40
010	69	50	9.6	0.3	F	25-30
012	70	51	8.2	0.3	F	25-30
013	70	50	10.2	0.3	F	25-30

* COI - cut-off index

Table S4: **The 50% serum neutralization titres against SARS-CoV-2 pseudoviruses**

Sample name	NT$_{50}$			% NT$_{50}$		
	Wuhan	Lineage B.1.1.7	Lineage B.1.351	Wuhan	Lineage B.1.1.7	Lineage B.1.351
003	237,3	272,3	47,95	100	114,75	20,21
004	89,63	197,5	59,94	100	220,35	66,87
005	110,6	211,7	35,47	100	191,41	32,07
006	526,8	172	132,4	100	32,65	25,13
008	310,4	144,2	56,87	100	46,46	18,32
010	234,5	152,9	36,97	100	65,20	15,77
012	239	111,4	68,15	100	46,61	28,51
013	289,6	161,3	34,83	100	55,70	12,03

The median 50% neutralization titers (NT50) of serum against the pseudo-virus SARS-CoV-2 Wuhan was 1/238, while the median NT50 for the pseudo-SARS-CoV-2 lineage of B.1.1.7 was 1/166. According to the one-way ANOVA test with Dunn's multiple comparisons, these differences were statistically insignificant. In testing the neutralizing activity of the serum against the SA variant of the pseudo-virus of the B.1.351 lineage a significant decrease in NT50 was observed compared to the variants of Wuhan and B.1.1.7 lineage ($p < 0.05$). The average NT50 for B.1.351 was 1/52, this is 75% lower than the value for the reference strain Wuhan.

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