Electronic Supplementary Material

Rapid Acquisition of High-Quality SARS-CoV-2 Genome via Amplicon-Oxford Nanopore Sequencing

Yi Yan[1,2,3,4,#] · Ke Wu[1,2,3,4,#] · Jun Chen[5,#] · Haizhou Liu[1,2,3] · Yi Huang[6] · Yong Zhang[1] · Jin Xiong[1] · Weipeng Quan[7] · Xin Wu[8] · Yu Liang[9] · Kunlun He[10,11] · Zhilong Jia[10,11] · Depeng Wang[8] · Di Liu[1,2,3,4,12✉] · Hongping Wei[1✉] · Jianjun Chen[1,2,✉]

1. CAS Key Laboratory of Special Pathogens and Biosafety, Wuhan Institute of Virology, Center for Biosafety Mega-Science, Chinese Academy of Sciences, Wuhan 430071, China
2. National Virus Resource Center, Wuhan Institute of Virology, Chinese Academy of Sciences, Wuhan 430071, China
3. Computational Virology Group, Center for Bacteria and Viruses Resources and Bioinformation, Wuhan Institute of Virology, Chinese Academy of Sciences, Wuhan 430071, China
4. University of Chinese Academy of Sciences, Beijing 101409, China
5. Wuhan Pulmonary Hospital, Wuhan Tuberculosis Prevention and Treatment Institute, Wuhan 430030, China
6. National Biosafety Laboratory, Chinese Academy of Sciences, Wuhan 430071, China
7. GrandOmics Biosciences, Wuhan 430000, China
8. GrandOmics Biosciences, Beijing 102200, China
9. GrandOmics Diagnostics, Wuhan 430000, China
10. Key Laboratory of Biomedical Engineering and Translational Medicine, Ministry of Industry and Information Technology, Chinese PLA General Hospital, Beijing 100039, China
11. Beijing Key Laboratory for Precision Medicine of Chronic Heart Failure, Chinese PLA General Hospital, Beijing 100039, China
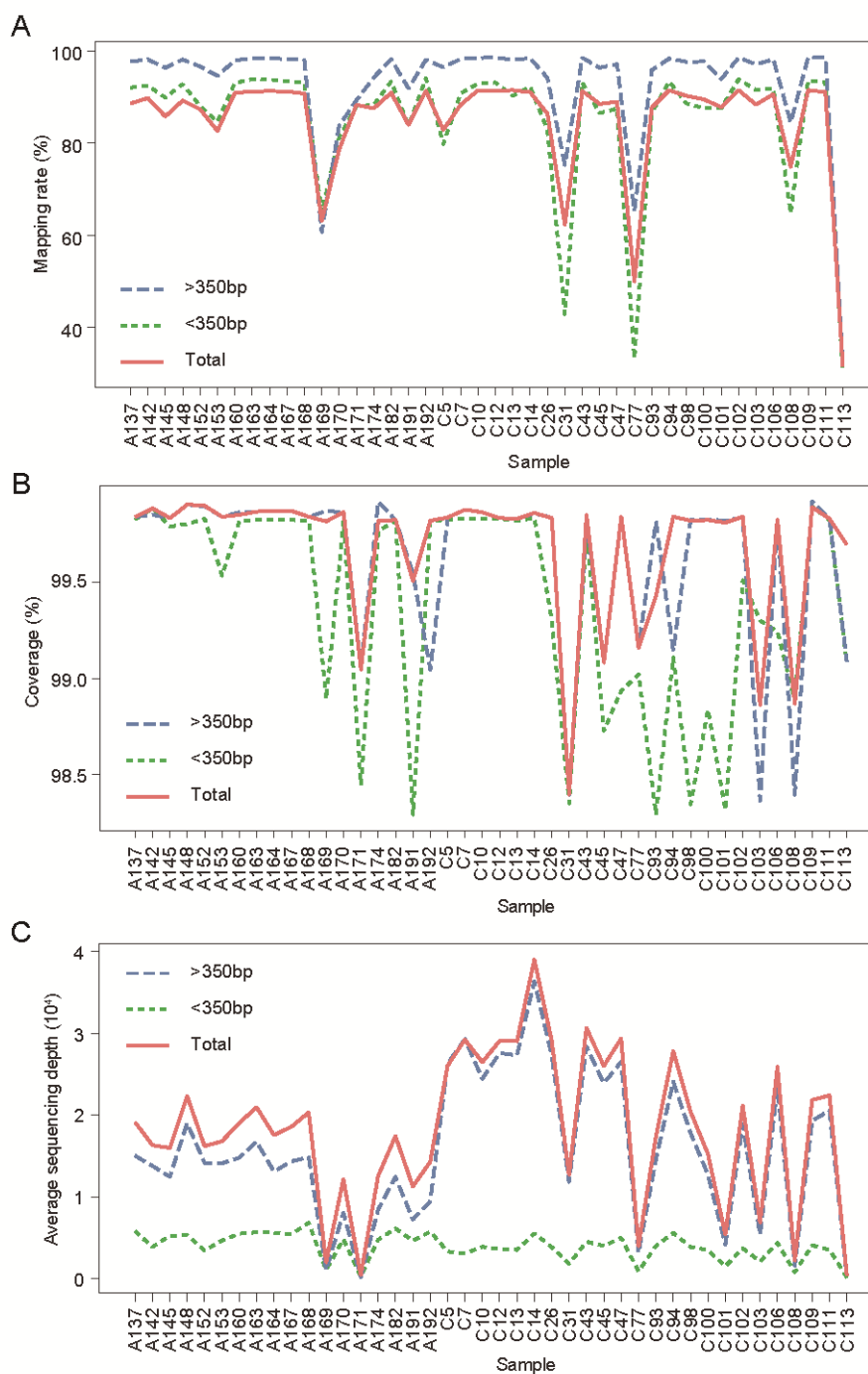12. First Affiliated Hospital of Xinjiang Medical University, Urumqi 830054, China

Fig. S1 Comparison of sequencing quality of SARS-CoV-2 between every data group. The proportion of reads mapped to the SARS-CoV-2 genome (**A**), the coverage of SARS-CoV-2 (**B**), and the average sequencing depth (**C**) of each sample in different data groups. Long-dashed line in blue represents data group only contained reads longer than 350 bp, dashed line in green represents data group only contained reads less than 350 bp, and solid line in red represents data group contained total high quality reads.
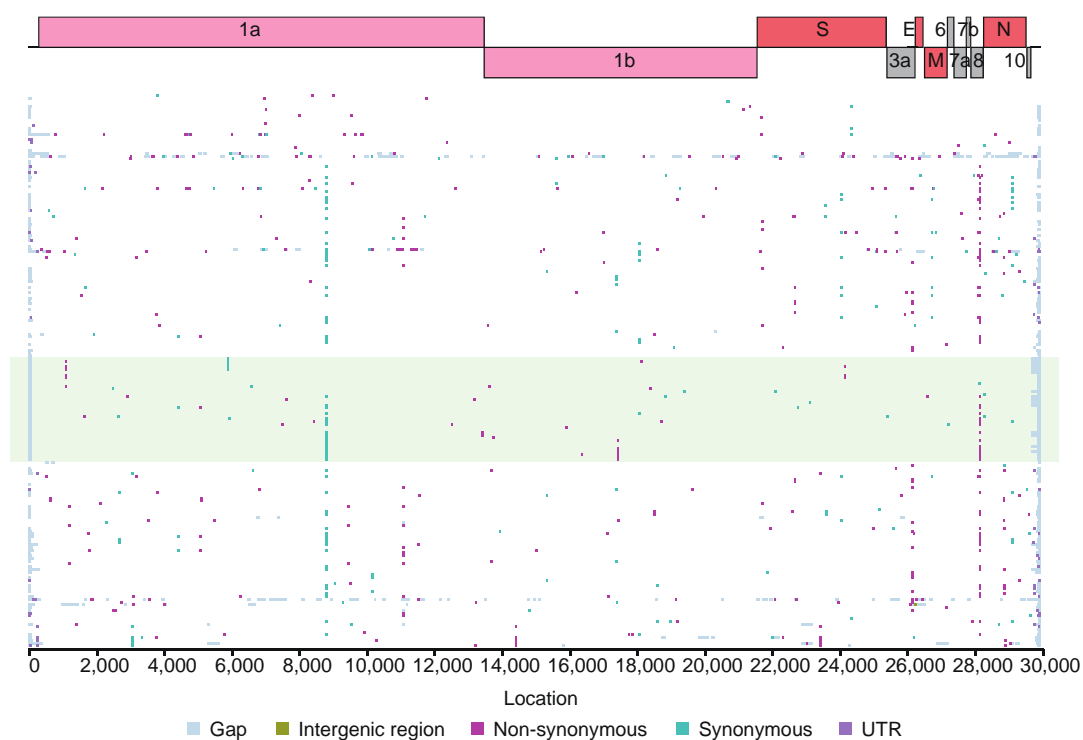
Fig. S2 Distribution of SNP sites in all released SARS-CoV-2 genome sequences until the submitted date of March 1, 2020. The upper part shows the open reading frames (ORFs) position of the reference genome, and the lower part displays the amino acid variations on the genome of each sample. Tiles in light grey are unmeasured regions in each genome, tiles in green are mutations in intergenic regions, tiles in blue represent synonymous mutations, tiles in red represent non-synonymous mutations, and tiles in purple represent mutations in UTR regions. All the strains were arranged from top to bottom according to the sampling time, and the light green background covered the 38 samples in this study.

Table S1 Sequencing data details for each sample.

Sample	Ct value	GISAID accession	NGDC accession	Total bases (Mb)	Total reads (M)	Mapping rate	Coverage	Average depth	Median depth	Batch
A137	23.6	EPI_ISL_493149	GWHALPE01000000	688.42	1.93	88.6	99.84	19,157.80	7,176.50	PAE36018
A142	20.83	EPI_ISL_493150	GWHALPF01000000	580.19	1.58	89.75	99.88	16,344.90	10,617.00	PAE36018
A145	24.57	EPI_ISL_493151	GWHALPG01000000	596.69	1.68	85.76	99.83	16,011.20	2,525.50	PAE36018
A148	21.99	EPI_ISL_493152	GWHALPH01000000	795.15	2.18	89.23	99.9	22,340.70	9,135.50	PAE36018
A152	27.88	EPI_ISL_493153	GWHALPI01000000	592.51	1.61	87.33	99.89	16,238.50	3,121.00	PAE36018
A153	27.2	EPI_ISL_493154	GWHALPJ01000000	648.73	1.79	82.63	99.84	16,815.40	2,074.00	PAE36018
A160	28.67	EPI_ISL_493155	GWHALPK01000000	664.69	1.87	90.88	99.85	19,063.10	15,387.00	PAE36018
A163	20.03	EPI_ISL_493156	GWHALPL01000000	725.14	2.02	91.18	99.86	20,985.60	19,474.50	PAE36018
A164	18.74	EPI_ISL_493157	GWHALPM01000000	607.94	1.73	91.27	99.87	17,588.40	16,320.00	PAE36018
A167	20.48	EPI_ISL_493158	GWHALPN01000000	642.66	1.82	91.18	99.87	18,594.60	17,253.50	PAE36018
A168	21.8	EPI_ISL_493159	GWHALPO01000000	707.23	2.02	90.73	99.84	20,350.80	19,057.00	PAE36018
A169[#]	35.92	EPI_ISL_493160	GWHALSE01000000	108.86	0.32	63.11	99.81	2,029.00	326	PAE36018
A170	31.992	EPI_ISL_493161	GWHALPP01000000	498.12	1.43	78.55	99.86	12,176.80	7,887.50	PAE36018
A171[#]	20.01	EPI_ISL_493162	GWHALSF01000000	18.59	0.06	88.28	99.05	472	185	PAE36018
A174	29.57	EPI_ISL_493163	GWHALPQ01000000	458.19	1.33	87.51	99.82	12,575.00	7,746.00	PAE36018
A182	27.69	EPI_ISL_493164	GWHALPR01000000	609.44	1.75	90.91	99.82	17,461.50	13,570.00	PAE36018
A191	29.41	EPI_ISL_493165	GWHALPS01000000	429.22	1.26	83.94	99.5	11,291.30	6,478.50	PAE36018
A192	25.734	EPI_ISL_493166	GWHALPT01000000	497.37	1.45	91.34	99.82	14,314.00	10,021.00	PAE36018
C5	31.77	EPI_ISL_493167	GWHALRI01000000	991.52	2.61	82.86	99.83	26,061.20	4,197.00	PAE38111
C7	27.17	EPI_ISL_493168	GWHALRJ01000000	1,046.89	2.74	88.32	99.87	29,253.90	10,587.00	PAE38111
C10	24.13	EPI_ISL_493169	GWHALRK01000000	912.12	2.44	91.42	99.86	26,503.40	24,644.00	PAE38111
C12	30.25	EPI_ISL_493170	GWHALRL01000000	1,002.25	2.65	91.36	99.83	29,106.50	27,562.00	PAE38111
C13	31.48	EPI_ISL_493171	GWHALRM01000000	1,003.46	2.66	91.46	99.83	29,055.90	24,545.00	PAE38111
C14	30.51	EPI_ISL_493172	GWHALRN01000000	1,352.64	3.60	91.02	99.86	39,045.60	35,678.00	PAE38111
C26	34.52	EPI_ISL_493173	GWHALRO01000000	1,064.40	2.83	86.34	99.83	29,137.80	22,198.50	PAE38111
C31	36.17	EPI_ISL_493174	GWHALRP01000000	642.85	1.73	62.32	98.4	12,738.30	6,782.00	PAE38111
C43	32.62	EPI_ISL_493175	GWHALRQ01000000	1,060.77	2.83	91.28	99.85	30,681.90	25,130.50	PAE38111
C45	35.3	EPI_ISL_493176	GWHALRR01000000	930.45	2.50	88.47	99.08	26,043.40	14,611.00	PAE38111
C47	34.47	EPI_ISL_493177	GWHALRS01000000	1,041.74	2.81	88.97	99.84	29,397.20	22,722.50	PAE38111
C77	39.14	EPI_ISL_493178	GWHALRT01000000	244.20	0.68	50.02	99.16	3,996.30	565	PAE38111
C93	32.08	EPI_ISL_493179	GWHALRU01000000	628.27	1.72	87.7	99.43	17,378.20	7,211.50	PAE38111
C94	30.42	EPI_ISL_493180	GWHALRV01000000	967.56	2.63	91.43	99.84	27,852.30	17,543.00	PAE38111
C98	31.06	EPI_ISL_493181	GWHALRW01000000	718.68	1.95	90.24	99.82	20,437.90	14,064.50	PAE38111
C100	31.13	EPI_ISL_493182	GWHALRX01000000	540.63	1.49	89.47	99.82	15,274.00	7,751.50	PAE38111
C101	34.62	EPI_ISL_493183	GWHALRY01000000	196.71	0.55	87.79	99.81	5,429.00	527	PAE38111
C102	27.26	EPI_ISL_493184	GWHALRZ01000000	729.65	1.96	91.46	99.84	21,167.90	18,023.00	PAE38111

C103	31.68	EPI_ISL_493185	GWHALSA01000000	249.47	0.69	88.35	98.86	6,906.10	441	PAE38111
C106	29.07	EPI_ISL_493186	GWHALSB01000000	901.70	2.42	90.61	99.82	25,918.90	20,317.50	PAE38111
C108[#]	37.66	EPI_ISL_493187	GWHALSG01000000	101.10	0.29	74.87	98.87	2,259.00	361	PAE38111
C109	27.33	EPI_ISL_493188	GWHALSC01000000	754.43	2.04	91.47	99.89	21,883.80	18,596.00	PAE38111
C111	26.96	EPI_ISL_493189	GWHALSD01000000	776.08	2.08	91.05	99.83	22,434.50	20,085.00	PAE38111
C113[#]	36.85	EPI_ISL_493190	GWHALSH01000000	46.43	0.14	31.6	99.69	418	210	PAE38111

Ct: Cycle threshold. [#]: Samples didn't satisfy the standard that more than 90% of genome were covered at same time with depth greater than 100×.

Table S2 Nucleotide variation site information

Mutation	ORF	Protein	Nucleotide substitution	n/s[a]	Amino acid substitution	Sample
1079*	ORF1a	NSP2	A-C	n	N-H	A142, A148, A512, A160, A163, A168
1620*	ORF1a	NSP2	A-C	n	E-A	C14
2462*	ORF1a	NSP2	C-T	s	-	A170
2623*	ORF1a	NSP2	C-T	s	-	C14
2888*	ORF1a	NSP3	G-T	n	V-L	A191
4402	ORF1a	NSP3	T-C	s	-	C10
5062	ORF1a	NSP3	G-T	n	L-F	C10
5869*	ORF1a	NSP3	C-T	s	-	A137, A142, A145, A148, A152
5905*	ORF1a	NSP3	T-C	s	-	C26
6556*	ORF1a	NSP3	T-C	s	-	A168
7482*	ORF1a	NSP3	C-T	n	S-L	C43
7615*	ORF1a	NSP3	G-A	n	W-*	A192
8432*	ORF1a	NSP3	G-T	n	E-*	C31
8782	ORF1a	NSP4	C-T	s	-	A191, C7, C10, C13, C26, C31, C43, C77, C93, C94, C98, C100, C101, C102, C103, C106, C109, C111
12513*	ORF1a	NSP8	C-T	n	T-M	C43
13175*	ORF1a	NSP10	A-G	n	T-A	A192
13402*	ORF1a	NSP10	T-G	n	Y-*	C77, C93
13625*	ORF1b	RdRp	A-G	n	D-G	A168
13730*	ORF1b	RdRp	C-T	n	A-V	C94

15907*	ORF1b	RdRp	G-A	n	G-S	C45
16349*	ORF1b	NSP13	C-T	n	S-L	C106
17410*	ORF1b	NSP13	C-T	n	R-C	C98, C102, C103, C106, C109, C111
18060	ORF1b	NSP14	C-T	s	-	C13
18109*	ORF1b	NSP15	C-A	n	P-T	A142
18380*	ORF1b	NSP16	G-A	n	G-D	A170
18712*	ORF1b	NSP17	G-T	n	A-S	C31
18816*	ORF1b	NSP18	G-T	s	-	A191
19386*	ORF1b	NSP14	C-T	s	-	A174
22081*	S	S	G-A	s	-	A174
22747*	S	S	C-T	s	-	C10
23116*	S	S	A-T	s	-	C5
24130*	S	S	C-A	n	N-K	A148, A160, A163
25383*	S	S	A-G	s	-	C14
26565*	M	M	A-C	n	K-Q	A191
27213	ORF6	ORF6	C-T	s	-	C43
28115*	ORF8	ORF8	C-T	s	-	A167
28144	ORF8	ORF8	T-C	n	L-S	A191, C7, C10, C13, C26, C31, C43, C77, C98, C101, C102, C103, C106, C109, C111
28253	ORF8	ORF8	C-T	s	-	A182, C14
29095	N	N	C-T	s	-	C31

[a] 'n' represents non-synonymous mutation and 's' represents synonymous mutation. * Newly detected nucleotide mutation sites.

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Table S3 Comparison of MuAFs between Nanopore and Illumina sequencing in 6 selected samples with more SNPs.

Mutation	A191 Nanopore	A191 Miseq	C10 Nanopore	C10 Miseq	C14 Nanopore	C14 Miseq	C31 Nanopore	C31 Miseq	C43 Nanopore	C43 Miseq	C106 Nanopore	C106 Miseq
A1548G	0.8377	0.9858	0.8199	0.9745	0.8197	0.9811	0.8262	0.9851	0.8196	0.9649	0.8195	0.9875
A1620C	NA	NA	NA	NA	0.8638	0.9795	NA	NA	NA	NA	NA	NA
C2623T	NA	NA	NA	NA	0.8318	0.9731	NA	NA	NA	NA	NA	NA
G2888T	0.7322	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
T4402C	NA	NA	0.7804	0.9801	NA	NA	NA	NA	NA	NA	NA	NA
G5062T	NA	NA	0.9422	0.995	NA	NA	NA	NA	NA	NA	NA	NA
C7482T	NA	NA	NA	NA	NA	NA	NA	NA	0.922	0.9985	NA	NA
G8432T	NA	NA	NA	NA	NA	NA	0.8677	NA	NA	NA	NA	NA
G8742T	NA	NA	NA	NA	NA	NA	NA	0.9726	NA	NA	NA	NA
T8782C	NA	NA	NA	NA	0.8965	0.9979	NA	NA	NA	NA	NA	NA
C12513T	NA	NA	NA	NA	NA	NA	NA	NA	0.937	0.9893	NA	NA
C16349T	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	0.9192	0.9715
C17410T	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	0.9324	0.9921
G18712T	NA	NA	NA	NA	NA	NA	0.8432	NA	NA	NA	NA	NA
G18816T	0.8302	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
C22747T	NA	NA	0.9443	0.9993	NA	NA	NA	NA	NA	NA	NA	NA
T24034C	0.8879	0.9995	0.882	0.9984	0.8836	0.998	0.9032	1	0.8855	0.9996	0.891	0.999
A25383G	NA	NA	NA	NA	0.8949	0.9577	NA	NA	NA	NA	NA	NA
A26565C	0.7409	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
C26729T	0.9192	0.9985	0.9006	0.9974	0.8942	0.9992	0.9026	0.9996	0.9024	0.999	0.899	0.9996
C27213T	NA	NA	NA	NA	NA	NA	NA	NA	0.613	0.6015	NA	NA
C28077G	0.9488	0.9903	0.9407	0.9846	0.9414	0.9846	0.9437	0.9057	0.9423	0.9846	0.9408	0.9907
C28144T	NA	NA	NA	NA	0.932	0.9895	NA	NA	NA	NA	NA	NA
C28253T	NA	NA	NA	NA	0.6146	0.6069	NA	NA	NA	NA	NA	NA
T28792A	0.9572	0.9797	0.9499	0.9726	0.9478	0.97	0.9492	0.9679	0.9464	0.974	0.9474	NA
C29095T	NA	NA	NA	NA	NA	NA	0.8524	0.9931	NA	NA	NA	NA
T29225C	NA	NA	NA	NA	NA	NA	NA	0.9963	NA	NA	NA	NA