

COVID-19 subject TWIST RNA

2021-05-21

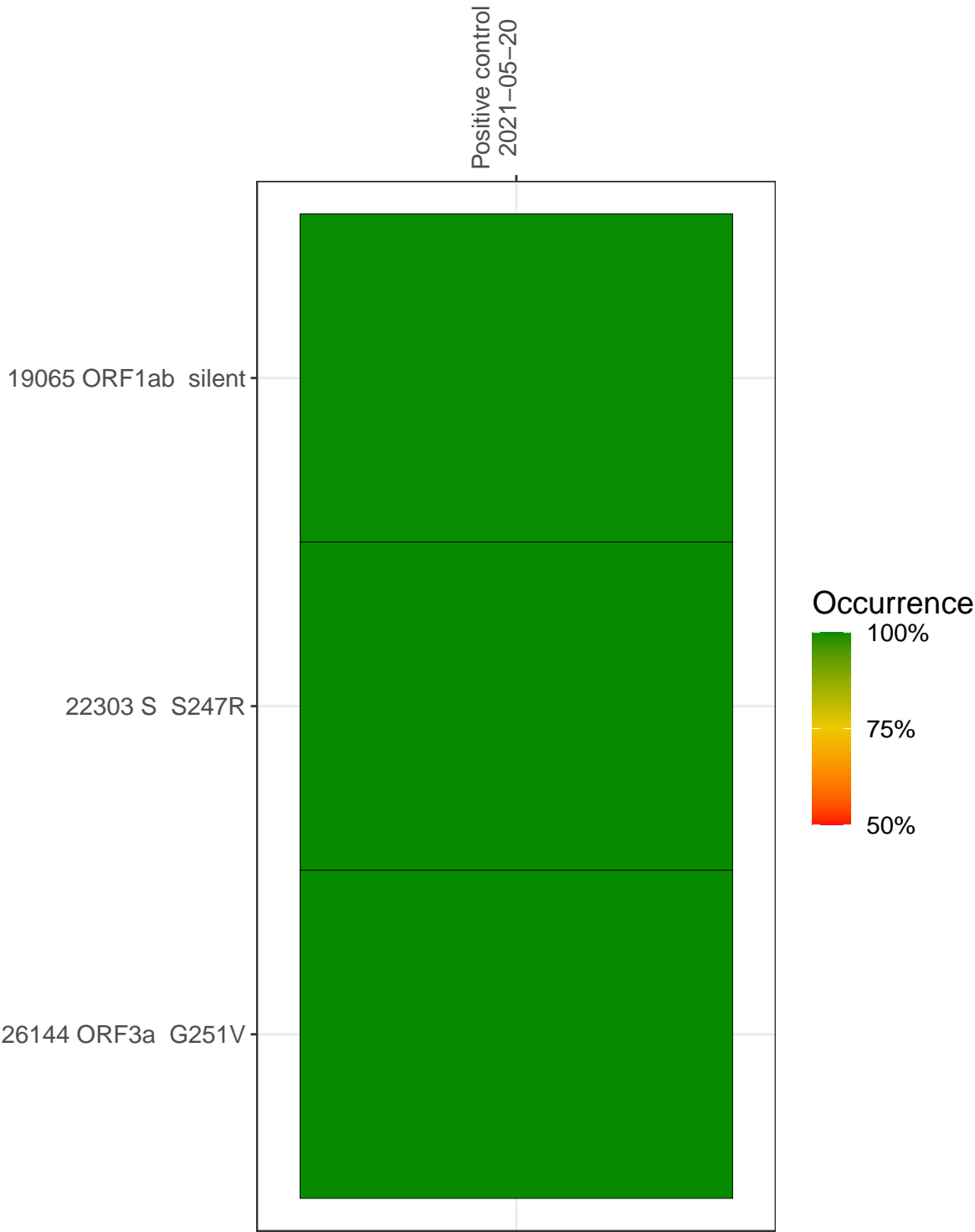
The table below provides a summary of subject samples for which sequencing data is available. The experiments column shows the number of sequencing experiments performed for each specimen. Experiment specific analyses are shown at the end of this report. Lineages are called with the Pangolin software tool (Rambaut et al 2020) for genomes with $> 90\%$ sequence coverage.

Table 1. Sample summary.

| Experiment | Type | Genomes | Sample type | Sample date | Largest contig (KD) | Lineage | Reference read coverage | Reference read coverage (≥ 5 reads) |
|------------|-------------------|---------|------------------|-------------|---------------------|---------|-------------------------|---|
| VSP3000 | composite | NA | Positive control | 2021-05-20 | 5.11 | B | 99.5% | 98.2% |
| VSP3000-1 | single experiment | NA | Positive control | 2021-05-20 | 4.99 | B | 98.2% | 96.2% |
| VSP3000-2 | single experiment | NA | Positive control | 2021-05-20 | 5.07 | B | 99.2% | 97.5% |
| VSP3000-3 | single experiment | NA | Positive control | 2021-05-20 | 5.01 | NA | 97.8% | 94.9% |

Variants shared across samples

The heat map below shows how variants (reference genome /home/everett/projects/SARS-CoV-2-Philadelphia/Wuhan-Hu-1) are shared across subject samples where the percent variance is colored. Variants are called if a variant position is covered by 5 or more reads, the alternative base is found in > 50% of read pairs and the variant yields a PHRED score > 20. Gray tiles denote positions where the variant was not the major variant or no variants were found. The relative base compositions of each experiment used to calculate tiles are shown in the following plot where the total number of position reads are shown atop of each plot.



Positive control
2021-05-20

19065 ORF1ab silent

6784

15751

1544

22303 S S247R

1157

3784

491

26144 ORF3a G251V

7173

14005

1560

VSP3000-1

VSP3000-2

VSP3000-3

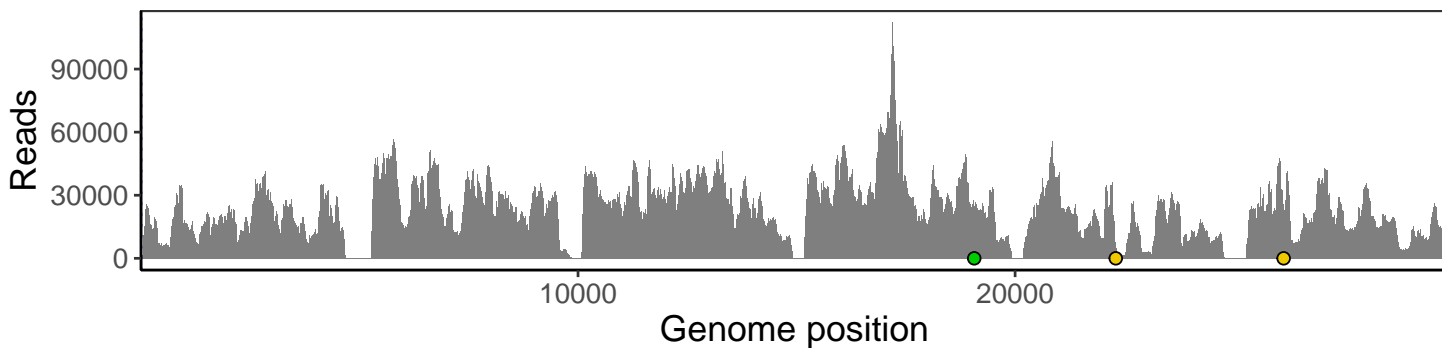
Base change

- Expected
- A
- T
- C
- G
- N
- Ins/Del
- No data

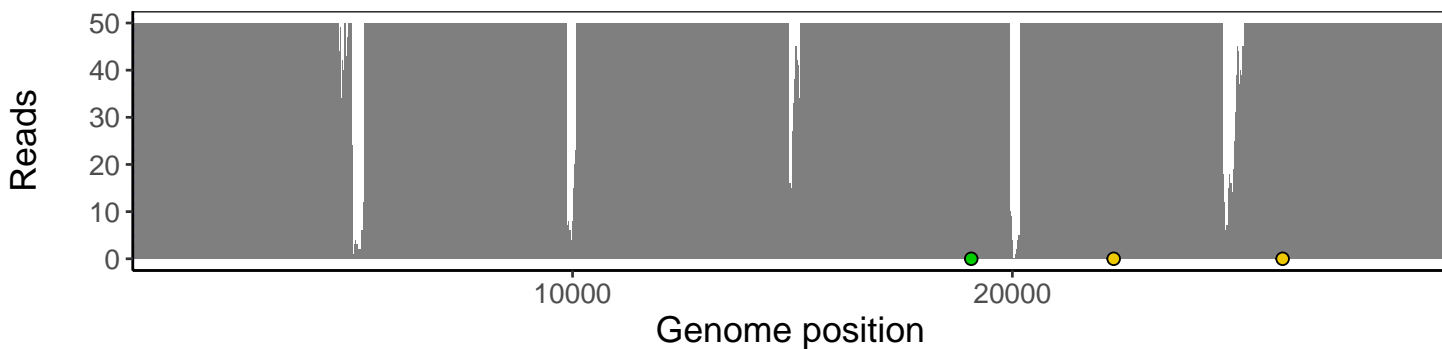
Analyses of individual experiments and composite results

VSP3000 | 2021-05-20 | Positive control | TWIST RNA | composite result

The plot below shows the number of reads covering each nucleotide position in the reference genome. Variants are shown as colored dots along the bottom of the plot and are color coded according to variant types: gray - transgenic, green - silent, gold - missense, red - nonsense, black - indel.



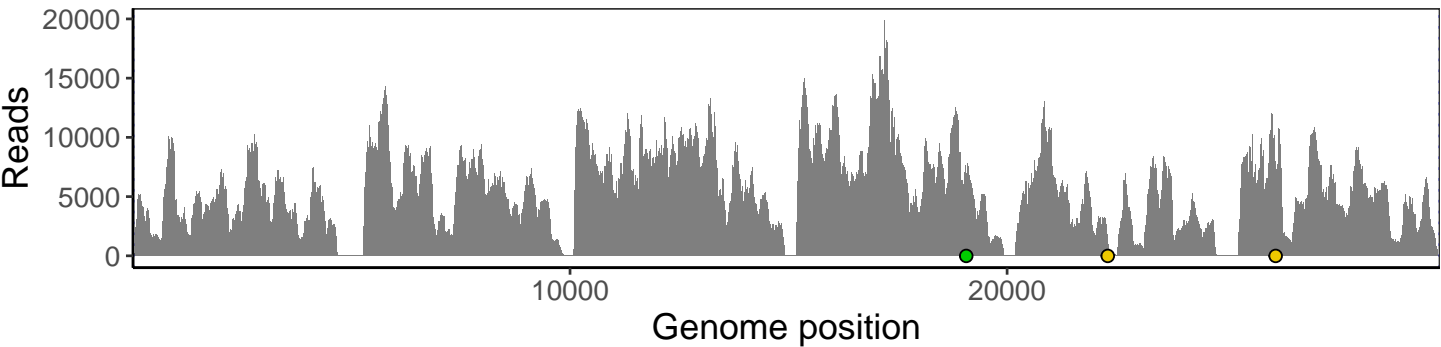
Excerpt from plot above focusing on reads coverage from 0 to 50 NT.



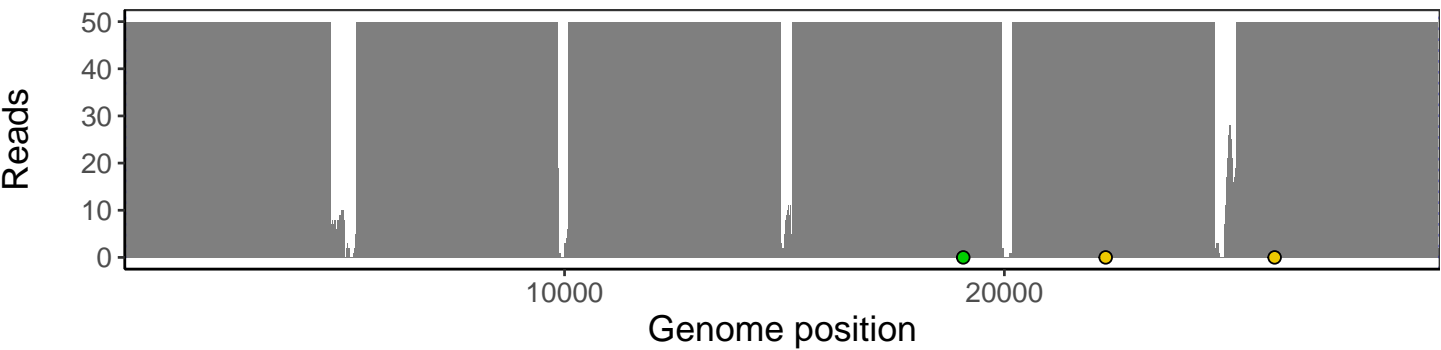
The longest five assembled contigs are shown below colored by their edit distance to the reference genome.



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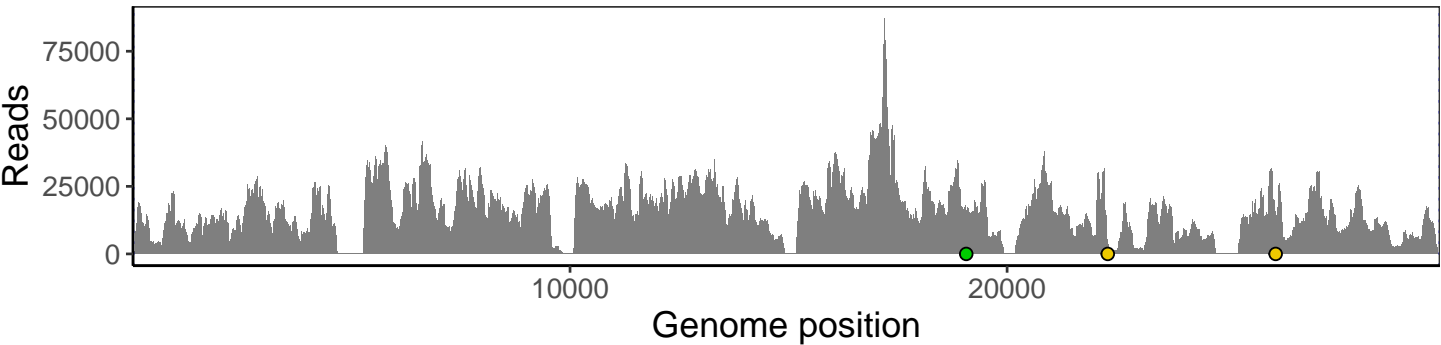
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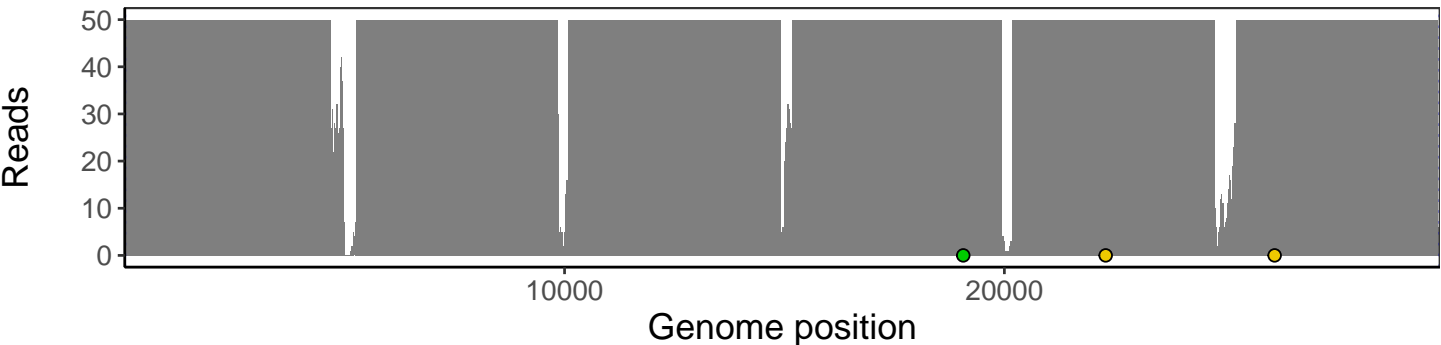
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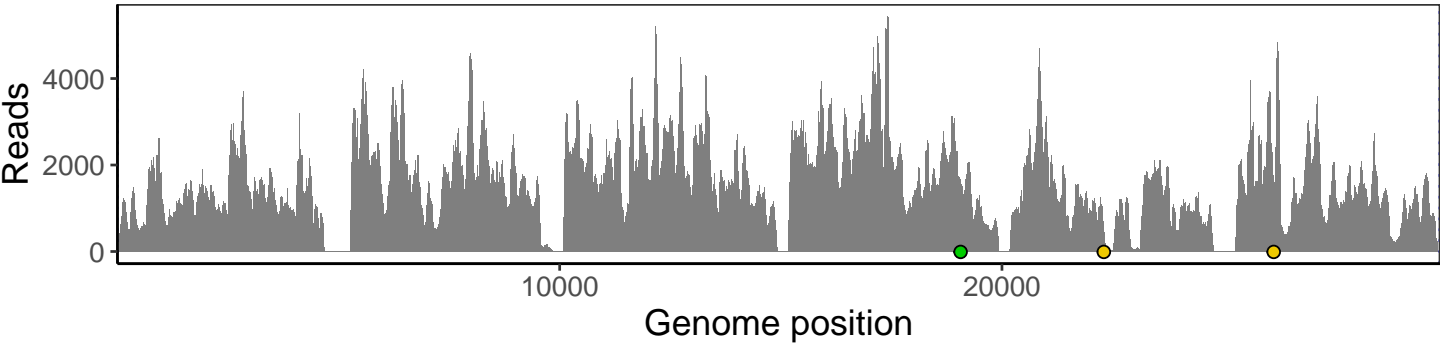
Excerpt from plot above focusing on reads coverage from 0 to 50 NT.



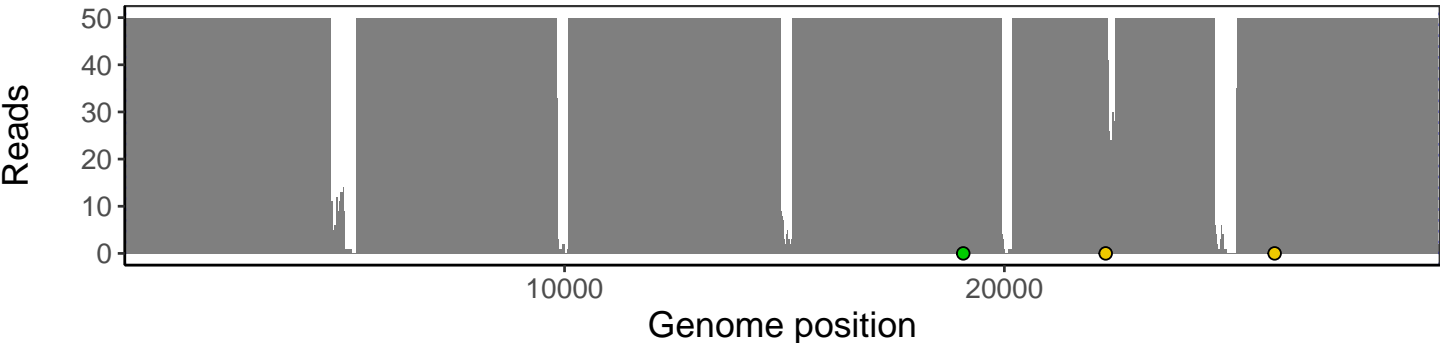
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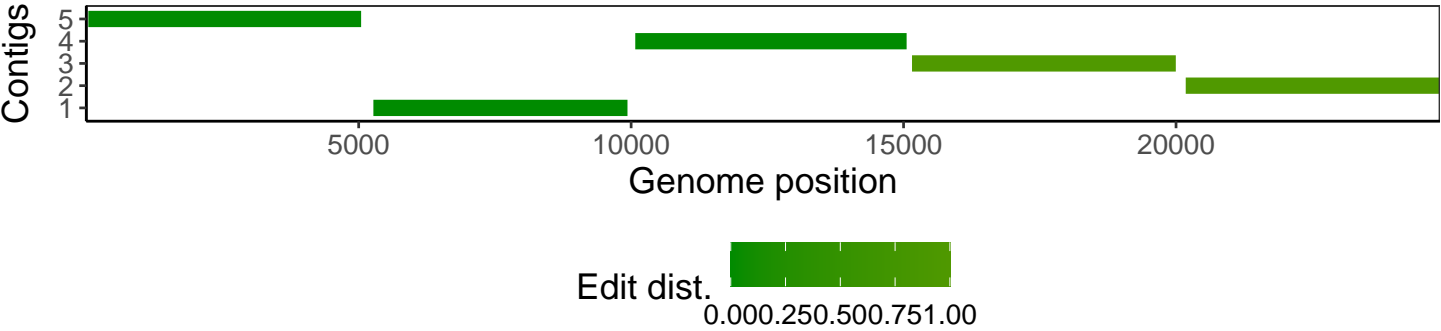
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Excerpt from plot above focusing on reads coverage from 0 to 50 NT.



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Software environment

| Software/R package | Version |
|----------------------|---|
| R | 3.4.0 |
| bwa | 0.7.17-r1198-dirty |
| samtools | 1.10 Using htlib 1.10 |
| bcftools | 1.10.2-34-g1a12af0-dirty Using htlib 1.10.2-57-gf58a6f3 |
| pangolin | 2.3.8 |
| genbankr | 1.4.0 |
| optparse | 1.6.0 |
| forcats | 0.3.0 |
| stringr | 1.4.0 |
| dplyr | 0.8.1 |
| purrr | 0.2.5 |
| readr | 1.1.1 |
| tidyr | 0.8.1 |
| tibble | 2.1.2 |
| ggplot2 | 3.3.3 |
| tidyverse | 1.2.1 |
| ShortRead | 1.34.2 |
| GenomicAlignments | 1.12.2 |
| SummarizedExperiment | 1.6.5 |
| DelayedArray | 0.2.7 |
| matrixStats | 0.54.0 |
| Biobase | 2.36.2 |
| Rsamtools | 1.28.0 |
| GenomicRanges | 1.28.6 |
| GenomeInfoDb | 1.12.3 |
| Biostrings | 2.44.2 |
| XVector | 0.16.0 |
| IRanges | 2.10.5 |
| S4Vectors | 0.14.7 |
| BiocParallel | 1.10.1 |
| BiocGenerics | 0.22.1 |