Report of SARS-CoV-2 B1.1.7 Lineage in Morocco

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ABSTRACT

Here, we report the near-complete genome sequence and the genetic variations of a clinical sample of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) harboring the N501Y mutation assigned to the B.1.1.7 lineage. The sample was collected from a nasopharyngeal swab of a female patient from Temara, Morocco, and the sequencing was done using Ion S5 technology.

A new Betacoronavirus strain of the Coronaviridae family named severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is the etiological agent of coronavirus disease 19 (COVID-19) (1–3). The identification of new mutations may contribute to characterizing the virus, mapping its spread, and better understanding its biological and clinical features (4, 5). In this report, near-whole-genome sequencing (WGS) of SARS-CoV-2 was carried out using Ion S5 sequencing technology to detect new variants (6).

The sampling was carried out on 8 January 2021. RNA was extracted from a nasopharyngeal swab sample of a 48-year-old female from Temara, Morocco, at the Central Laboratory of Virology, Hospital of Specialties of Rabat, using a Maxwell RSC blood DNA extraction kit (Promega, USA). The patient was identified as positive for COVID-19 by reverse transcriptase quantitative PCR using a SARS-CoV-2 kit (MAScIR, Morocco) and exhibited cycle threshold (Ct) values of 19 for both S and RdRp genes. The cDNA was prepared using a SuperScript VILO cDNA synthesis kit (Invitrogen, Thermo Fisher Scientific, USA). A total of 15 ml of cDNA was used to prepare a SARS-CoV-2 library by using an Ion AmpliSeq kit for Chef DL8 (Thermo Fisher Scientific, USA). The library was adjusted to 30 pM and loaded onto the Ion Chef instrument (Thermo Fisher Scientific, USA) for emulsion PCR, enrichment, and loading onto the Ion S5 530 chip. WGS was performed using the Ion AmpliSeq SARS-CoV-2 research panel designed by Thermo Fisher Scientific for complete viral genome sequencing according to instructions for use on an Ion Gene Studio S5 Prime series system.

Raw data were analyzed using Torrent Suite software v 5.12.0. The NGS QC Toolkit v 2.3.3 was used to remove low-quality and short reads. Variant Caller v 5.10.1.19 was used to detect variants compared to the reference genome (Wuhan-Hu-1, GenBank accession number MN908947.3), while the consensus sequence was generated using IRMAreport v 1.3.0.2. The annotation was carried out using COVID19AnnotateSnpEff v 1.3.0.2, a plugin specifically developed for SARS-CoV-2. Default parameters were used for all software (7).

Our analysis allowed us to obtain a near-complete SARS-CoV-2 genome of 29,805 bp length with an average read length of 206 bp and an overall DNA G+C content of 37.98%. From 879,763 reads, 862,414 reads were correctly mapped, covering 97.56% of the total genome with a mean depth of 5,726×.

The genetic variant process revealed a total of 34 variations, including 15 in open reading frame 1ab (ORF1ab; 7 synonymous variants, 6 missense variants, 1 conservative in-frame...
TABLE 1 Types and effects of identified gene variations compared to the reference strain, Wuhan-Hu-1 (GenBank accession number MN908947.3)

| Gene   | Nucleotide position | Nucleotide change | Residue change | Effect                       |
|--------|---------------------|-------------------|----------------|------------------------------|
| ORF1a  | c.−25C>T            | No change assigned| Upstream gene variant |
| 913    | c.648C>T            | p.Ser216Ser       | Synonymous variant |
| 3037   | c.2772C>T           | p.Phe924Phe       | Synonymous variant |
| 3267   | c.3002C>T           | p.Thr1001Ile      | Missense variant |
| 5388   | c.5123C>A           | p.Ala1708Asp      | Missense variant |
| 5986   | c.5721C>T           | p.Phe1907Phe      | Synonymous variant |
| 6954   | c.6689T>C           | p.Ile2230Thr      | Missense variant |
| 10277  | c.10012C>T          | p.Leu3338Phe      | Missense variant |
| 11287  | c.11023_11031delTCTGGTTTT | p.Ser3675_Phe3677del | Conservative in-frame deletion |
| 14408  | c.14144C>T          | p.Pro4715Leu      | Missense variant |
| 14676  | c.14412C>T          | p.Pro4804Pro      | Missense variant |
| 14925  | c.14661C>T          | p.Val4887Val      | Missense variant |
| 15279  | c.15015C>T          | p.His5005His      | Missense variant |
| 16176  | c.15912T>C          | pThr5304Thr      | Synonymous variant |
| 17615  | c.17351A>G          | p.Lys5784Arg      | Missense variant |
| S      | c.204_209delACATGT  | p.His69_Val70del | Disruptive in-frame deletion |
| 21990  | c.432_434delTTTA    | p.Tyr145del       | Disruptive in-frame deletion |
| 23063  | c.1501A>T           | p.Asn501Tyr       | Missense variant |
| 23271  | c.1709C>A           | p.Ala570Asp       | Missense variant |
| 23403  | c.1841A>G           | p.Asp614Gly       | Missense variant |
| 23604  | c.2042C>A           | p.Pro681His       | Missense variant |
| 23709  | c.2147C>T           | p.Thr716Ile       | Missense variant |
| 24506  | c.2944T>G           | p.Ser982Ala       | Missense variant |
| 24914  | c.3352G>C           | p.Asp1118His      | Missense variant |
| ORF8   | c.79C>T             | p.Gln27*          | Stop gained     |
| 28048  | c.155G>T            | p.Arg52Ile        | Missense variant |
| 28111  | c.218A>G            | p.Tyr73Cys        | Missense variant |
| N      | c.7G>C              | p.Asp3His         | Missense variant |
| 28281  | c.8A>T              | p.Asp3Val         | Missense variant |
| 28282  | c.9T>A              | p.Asp3Glu         | Missense variant |
| 28881  | c.608G>A            | p.Arg203Lys       | Missense variant |
| 28882  | c.609G>A            | p.Arg203Arg       | Synonymous variant |
| 28883  | c.610G>C            | p.Gly204Arg       | Missense variant |
| 28977  | c.704C>T            | p.Ser235Phe       | Missense variant |

* A stop codon.

deletion, and 1 upstream gene variant), 9 in spike genes (7 missense variants and 2 disruptive in-frame deletions), 3 in ORF8 (2 missense variants and 1 stop gained), and 7 in the N gene (6 missense variants and 1 synonymous variant). The spike gene carries the mutation known as N501Y (Asn501Tyr; c.1501A>T). This mutation cooccurs with several mutations, including missense mutations (A570D, P681H, T716I, S982A, and D1118H), as well as disruptive in-frame deletions (H69-V70 and Y145) (8, 9). The genomic features of the sequenced sample are summarized in Table 1. The phylogenetic analysis using Phylogenetic Assignment of Named Global Outbreak (PANGO) lineages (10) revealed that the strain belongs to the B1.1.7 lineage.

Data availability. This sequence was deposited in the GenBank and GISAID databases under the accession numbers MW803167 and EPI_ISL_1137621, respectively. The raw reads were deposited in the NCBI Sequence Read Archive (SRA) under the accession number SRR13811335.

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We declare no competing interests.

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