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Detection of the SARS-CoV-2 UK Variant in Portugal †

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Abstract: At the end of 2020, a new highly transmissible variant of SARS-CoV-2 was discovered in the United Kingdom (UK). This work aims to identify potential cases of the UK variant in Portugal using routine diagnostic samples. A total of 26 out of 43 positive samples that were identified by RT-PCR as suspects were confirmed through sequencing to be the SARS-CoV-2 UK variant. The first case of the UK variant identified by us was in samples collected on 21 December 2020 at Lisbon airport in travelers from Manchester and London.

Keywords: SARS-CoV-2; COVID-19; Genomic epidemiology; UK variant; Spike mutation

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1. Introduction

The SARS-CoV-2 strain lineage B.1.1.7, also known as the UK variant (or 501Y.V1), was identified as highly contagious in the UK. This variant, with the potential ability to evade host immunity [1,2], creates new challenges in the control of the pandemic, making the detection and tracking of this variant and others alike crucial to the long-term containment of SARS-CoV-2, mostly in the context of mass-vaccination. The early identification of B.1.1.7 in patients may help reducing further spread of this variant.

B.1.1.7 has a specific mutation in the Spike(S)-gene, which results in a deletion of two amino acids at sites 69 (histidine) and 70 (valine) (69–70 del) [3]. The multi-target design of some diagnostic tests that includes an S-gene target may function as a preliminary screening for the presence of mutations in S-gene, being a first clue of the B.1.1.7 lineage. Nevertheless, a confirmation is required by genome sequencing to achieve accurate results.

The sequencing of all SARS-CoV-2-positive cases is an expensive and time-consuming technique that requires samples with a high concentration of viral RNA and a high degree of purity to get satisfactory results. Public and private laboratories can play an important role in the early identification of B.1.1.7 or others by making an initial screening of the positive samples based in a simple RT-PCR diagnostic test and signaling the suspects for immediate confirmation. In this work, all the samples that had shown an absence of the S-gene in TaqPath COVID-19 diagnostic tests were considered as potentially being the UK variant.

2. Materials and Methods

Samples were obtained from routine diagnostics at SYNLAB Lisbon Laboratory. RNA was extracted from nasopharyngeal swabs and SARs-CoV-2 RNA detection was carried out through a multiplex real-time RT-PCR. Between December 21 (2020) and January 10 (2021), all positive samples, collected on the arrival area at International Lisbon Airport, were tested with a triple target assay, including S-gene target (Applied Biosystems TaqPath COVID-19 CE-IVD RT-PCR Kit), according to the manufacturer's instructions. In cases where S-gene was not detected but ORF1ab and N genes were amplified, RNA samples

were sequenced and analyzed by the National Institute of Health (INSA) Doctor Ricardo Jorge, Portugal. The UK variant was also analyzed in the community using the same approach in random samples collected between 6 to 9 of January. Some of them were also sequenced.

This study was performed in accordance with the General-Directorate of Health (DGS) guidelines. Informed consent was obtained from all patients and all samples are obtained under the service-providing activity of the SYNLAB Health laboratory in the diagnostic area, and thus, the ethical procedures inherent in obtaining them are fully respected.

3. Results and Discussion

A screening of a total of 76 cases of SARS-CoV-2-positive travelers is summarized in Table 1.

Table 1. Identification of SARS-CoV-2 UK variant in travelers that arrived at Lisbon airport.

Positive Cases	S gene Dropout	Identified as S: N501Y.V1 UK
76	43 (56.6%)	26 (60.5%) [1]

[1] 17 cases not identified as 501Y.V1 without any NGS result associated.

Most of the analyzed samples were from British travelers. In total, 60.5% of the SARS-CoV-2-positive cases with S-gene dropout were confirmed by sequencing as being the UK variant (lineage B.1.1.7). It should be noted that of the 43 S-gene dropout samples, 17 could not be sequenced because of the RNA quality needed for sequencing. Curiously, one of the confirmed UK variant samples was identified in a traveler who departed from Chisinau, Moldova International Airport.

Two weeks after the first cases were identified, when applying the same methodology to random cases detected as positive in the community samples, we found that 11 of the 13 SARS-CoV-2 infection cases that showed S-gene dropout after being sequenced proved to be the UK variant. These samples were collected in Lisbon, Setubal, and Algarve. This suggests that the UK variant had a fast spread within the community. At this time, the lab screened around 558 samples for S-gene detection per day and it was observed that from 148 $\pm$ 33 positive cases, 16 $\pm$ 3% did not amplify the S-gene.

Given the high number of positive samples that need to be screened through genome sequencing to identify the UK variant, this work demonstrates the importance of using RT-PCR diagnostic tests in diagnostic laboratories as a preliminary screening tool for early signaling of potential UK variant cases before genome sequencing. This work also demonstrates the important role of private diagnostic laboratories in complementing the Public Institutions regarding the identification of highly spreading variants.

Institutional Review Board Statement: Ethical review and approval were waived for this study, due to the fact that this study was based on laboratory routine results of Covid detection and performed according the demand of national authorities of health in a pandemic context.

Informed Consent Statement: Informed consent was obtained from all subjects involved in the study.

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Conflicts of Interest: The authors declare no conflict of interest.

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