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FULL GENOME SEQUENCE OF THE FIRST SARS-COV-2 ISOLATES DETECTED IN THE REPUBLIC OF MOLDOVA

Mariana ULINICI

Nicolae Testemitanu State University of Medicine and Pharmacy, Republic of Moldova

Corresponding author: Mariana Ulinici, e-mail: mariana.ulinici@usmf.md

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Introduction. Since the COVID-19 outbreak in late December 2019, more than 290 043 cases and 6713 deaths have been reported in the Republic of Moldova as of 27th of September 2021. The paucity of in-depth knowledge of the risk factors for severe COVID-19, insufficient diagnostic tools for the detection of SARS-CoV-2, absence of specific and effective drug treatments as well as the absence of a best measure to monitor emerging pathogens like SARS-CoV-2 led to a major burden on health care and economic systems across the world. As SARS-CoV-2 continues to acquire genetic changes over time during this pandemic, the full genome sequencing and identification of viral variants must continue.

This study aimed to perform whole-genome sequencing of SARS-VoV-2 isolates from Moldova and to provide a 'real-time' overview of the viral genotypes circulating in this geographical area. Suspected cases were screened for SARS-CoV-2 as per the advisory of the Ministry of Health.

Material and methods. 4 nasopharyngeal swabs were collected in June 2020. Samples were obtained from patients aged between 20 and 60 years old, residing in the Republic of Moldova. All samples from COVID-19 patients were collected from individuals with mild and severe COVID-19 symptoms and confirmed by real-time reverse transcription-polymerase chain reaction targeting the envelop (E) and nucleocapsid (N). The RNA samples were sequenced using an Illumina MiSeq sequencer according to standard procedure developed previously. The metadata was created and the sequences were deposited on GISAID international repository.

Results. Four patients from different geographic regions of the Republic of Moldova were confirmed positive for SARS-CoV-2. Complete (29,900 nucleotides, 29,886 nucleotides, 27,179 nucleotides, 29,867 nucleotides) genomes were obtained. Phylogenetic analysis showed that the Moldovan sequences belonged to different clusters. EPI_ISL_516935 (GISAID accesion ID) belongs to clade G and EPI_ISL_516934, EPI_ISL_516936, EPI_ISL_516938 belong to clade GR. These clades are prevalent in EU. All isolates showed typical mutation (Spike D614G) for G clade. Three genomes showed mutations on nucleocapsid protein, the G204R which is specific for GR clade. We also observed other amino-acids substitutions (NS3 V90F, NSP2 G339S, NSP3 D218E, NSP3 V1243A, NSP12 P323L, Spike E1202Z, Spike L763V, NS7a E95Z, NSP3 N1785D, Spike G769V, N P151L, N R203K, NSP12 R457C, NSP13 A379V, NSP15 N4S NS3 S165F, NSP2 L410F), mutations that are less frequent.

Conclusions. The four SARS-CoV-2 sequences obtained from Republic of Moldova represent two different introductions into the country. The genetic heterogeneity is as noted globally. The Spike D614G and N G204R has a rapid spread in the pandemic. Continuous monitoring and analysis of the sequences of new cases from Republic of Moldova and the other affected countries would be vital to understand the genetic evolution and rates of substitution of the SARS-CoV-2.