

# Partial Sanger Sequence Analysis of SARS-CoV-2 Spike (S) Gene Detects the Delta and Alpha SARS-CoV-2 Variants of Concern for the First Time in Vihiga County

*This is a preliminary report prepared by CDC Kenya's Diagnostic Laboratory Systems Program and the KEMRI Centre for Global Health Research on June 11, 2021.*

## Summary of Findings

- The Sanger partial sequencing method provides a quick, low-cost pre-screening approach to rapidly identify variants of concern (VOC) or interest (VOI).
- Nineteen (19) SARS-CoV-2 RT-PCR positive samples from five sub counties within Vihiga County were sequenced including: Emuhaya (3), Hamisi (1), Luanda (2), Sabatia (5) and Vihiga (8). These samples were collected between May 31 - June 8, 2021 from patients aged 18-81 years; 7 (37%) were female and 11 (58%) male. Two cases did not indicate sex or age on their case investigation form (CIF). All cases were Kenyan nationals, and none of the 19 cases indicated a recent travel history on their CIF (i.e., travel within the past 14 days).
- The following VOC were identified:
  - **11 samples (57.9%) aligned with the Delta SARS-CoV-2 VOC (B.1.617.2 first identified in India);** and
  - 8 samples (42.1%) aligned with the Alpha SARS-CoV-2 VOC (B.1.1.7 first identified in the United Kingdom).
- **This is the first time we have detected the Alpha and Delta SARS-CoV-2 VOCs in Vihiga county. The Delta VOC was detected in four of the five sub counties of Vihiga.**
- Representatives of the identified variants will undergo whole genome sequencing to confirm the assigned lineages.

## Background

There is increasing need for genomic surveillance of SARS-CoV-2 to determine new introductions and current circulation of variants of concern (VOC) and variants of interest (VOI) in the local population. Timely release of sequencing data is important for initiating mitigation measures and linking variants to clinical manifestation and vaccine effectiveness in our population.

Here we describe findings from partial S gene sequencing at the CDC/KEMRI laboratory in Kisumu of 19 SARS-CoV-2 samples collected from Vihiga county. These samples were collected between May 31 through June 8, 2021.

## Methods

Portions of spike (S) gene at nucleotide positions 21358 to 23847 of SARS-CoV-2 reference sequence (NC\_045512) genome were amplified using Qiagen Onestep RT-PCR kit, and nested PCR was performed using New England BioLabs Taq DNA polymerase. The PCR products were purified using ExoSAP-IT (ThermoFisher) and cycle sequencing was performed using BigDye sequencing kit v3.1. Excess Bigdye removal was done using DyeEx 96 kit (Qiagen) and resultant purified products read-out in ABI 3730xl genetic sequence analyser. Multiple sequence alignment (MSA) was generated using BioEdit v7.2. Maximum likelihood (ML) tree was constructed in MEGA v7.

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## Results - Sequence and Phylogenetic Analysis

All the 11 samples (57.9%) that clustered with the sub-lineage B.1.617.2 (Delta SARS CoV-2) contained amino acid changes at L452, T478K and D614G. Of these eight (72.7%) had amino acid changes at G142D and six (54.5%) had deletions at amino acid positions 156 and 157. All the eight samples that aligned with B.1.1.7 (Alpha SARS CoV-2) had deletions at amino acid 69/70 and 144. Amino acid changes at N501Y, A570D and D614G found in this lineage were also identified in these samples. Of the samples that aligned with Alpha VOC, five (62.5%) had amino acid change F490S and three (37.5%) had amino acid change D138H.

Information including age, sex, location and cluster are shown in the table below.

## Conclusions

- This report provides preliminary sequencing results that identified the Delta VOC (B.1.617.2) and Alpha VOC (B.1.1.7) in Vihiga County for the first time. These findings may inform public health action in the county.
- Using Sanger partial sequencing of the SARS-CoV-2 S gene, we identified 11 cases (57.9%) of the Delta VOC (B.1.617.2) in Vihiga County between May 31 - June 8, 2021. All cases were Kenyan nationals with no history of travel reported on their CIF.
- We also identified eight cases (42.1%) with the Alpha VOC (B.1.1.7) for the first time in Vihiga County.
- All results from this report require further confirmation with whole genome sequencing to confirm the lineages identified by partial sequencing.

## Acknowledgement

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**Table. Case information of sequences identified in five sub-counties of Vihiga County who presented at the Vihiga County Referral Hospital**

Age (Years)	Gender	Sub County	Symptoms	Date of sample collection	Pango lineage	WHO lineage
42	M	Emuhaya	No	6/3/2021	B.1.1.7	Alpha
78	F		Yes	6/7/2021	B.1.617.2	Delta
65	M		No	6/7/2021	B.1.617.2	Delta
66	F	Hamisi	Yes	6/8/2021	B.1.1.7	Alpha
27	F	Luanda	Yes	6/3/2021	B.1.1.7	Alpha
38	F		Yes	6/4/2021	B.1.617.2	Delta
81	F	Sabatia	Yes	6/2/2021	B.1.617.2	Delta
71	M		Yes	6/2/2021	B.1.617.2	Delta
75	M		Yes	6/3/2021	B.1.617.2	Delta
47	F		No	6/3/2021	B.1.617.2	Delta
77	M		Yes	6/7/2021	B.1.617.2	Delta
63	M	Vihiga	Yes	5/31/2021	B.1.1.7	Alpha
69	M		Yes	5/31/2021	B.1.1.7	Alpha
50	M		Yes	6/2/2021	B.1.1.7	Alpha
62	M		Yes	6/7/2021	B.1.1.7	Alpha
64	M		Yes	6/8/2021	B.1.1.7	Alpha
40	F		Yes	6/7/2021	B.1.617.2	Delta
NA	M		No	6/7/2021	B.1.617.2	Delta
18	NA		Yes	6/7/2021	B.1.617.2	Delta

Note: No cases reported a travel history within the last 14 days from the date of sample collection; NA = not available