

Supplemental Methods

V3 primer set modifications and PCR

Modified V3 pool1-M was generated by mixing 5 μ l of each primer nCoV-2019_5_RIGHT, nCoV-2019_5_LEFT, nCoV-2019_9_RIGHT, nCoV-2019_9_LEFT, nCoV-2019_17_RIGHT, nCoV-2019_17_LEFT, nCoV-2019_23_RIGHT, nCoV-2019_23_LEFT, nCoV-2019_45_RIGHT, nCoV-2019_45_LEFT, nCoV-2019_55_RIGHT, nCoV-2019_55_LEFT, nCoV-2019_67_RIGHT, nCoV-2019_67_LEFT, nCoV-2019_71_RIGHT, nCoV-2019_71_LEFT, nCoV-2019_73_RIGHT, nCoV-2019_73_LEFT, nCoV-2019_91_RIGHT, nCoV-2019_91_LEFT, nCoV-2019_97_RIGHT and nCoV-2019_97_LEFT.

Modified V3 pool2-M was generated by mixing 5 μ l of each primer nCoV-2019_8_RIGHT, nCoV-2019_8_LEFT, nCoV-2019_10_RIGHT, nCoV-2019_10_LEFT, nCoV-2019_12_RIGHT, nCoV-2019_12_LEFT, nCoV-2019_20_RIGHT, nCoV-2019_20_LEFT, nCoV-2019_28_RIGHT, nCoV-2019_28_LEFT, nCoV-2019_50_RIGHT, nCoV-2019_50_LEFT, nCoV-2019_54_RIGHT, nCoV-2019_54_LEFT, nCoV-2019_66_RIGHT, nCoV-2019_66_LEFT, nCoV-2019_70_RIGHT, nCoV-2019_70_LEFT, nCoV-2019_74_RIGHT, nCoV-2019_74_LEFT, nCoV-2019_92_RIGHT and nCoV-2019_92_LEFT. In addition, pool 2-M also contained 7 μ l of primers nCoV-2019_64_RIGHT, nCoV-2019_64_LEFT and 6 μ l of primers nCoV-2019_98_RIGHT, nCoV-2019_98_LEFT.

Pool PCR #1 was setup using 2.5 μ l cDNA, 3.4 μ l V3 pool1 primers (10 μ M), 0.8 μ l V3 pool1-M primers (10 μ M), 12.45 μ l Q5 High-Fidelity 2X Master Mix (NEB, Ipswich, MA, USA) and 5.85 μ l dH₂O. Pool PCR #2 was setup using 2.5 μ l cDNA, 3.4 μ l V3 pool2 primers (10 μ M), 1 μ l V3 pool2-M primers (10 μ M), 12.5 μ l Q5 High-Fidelity 2X Master Mix and 5.6 μ l dH₂O was added.