Supplemental Material

Table of Contents	Page
Supplemental Methods	**2**
Supplemental Results	**3**
Supplemental Figure 1: RT-PCR Cycle threshold (Ct) values per specimen type over time from end-stage renal disease (ESRD) participants on maintenance dialysis with SARS-CoV-2 infection (N=59 specimens)	**4**
Supplemental Figure 2: Detection of SARS-CoV-2 IgM, IgG and IgA antibody titers among participants with positive antibodies (N=13) since diagnosis	**5**
References	**7**

Supplemental Methods

Specimen collection and testing

OP and AN specimens were collected using Flocked Swabs (COPAN Diagnostics, California, USA) and placed in viral transport media containers (Becton, Dickson and Company, New Jersey, USA). Saliva was collected in OMNIGene® ORAL kits (DNA Genotek, Ottawa, Ontario, Canada). Blood was collected in K2 EDTA tubes (Becton, Dickson and Company, New Jersey, USA). All specimens were stored at 2–8°C during transport. RT-PCR was performed on all AN, OP and SA specimens using the CDC 2019-Novel Coronavirus RT-PCR Diagnostic Panel for the detection of SARS-CoV-2 RNA (1). OP and AN specimens with a positive RT-PCR result of a cycle threshold (Ct) of ≤34 were kept at -70°C and submitted for viral culture. Because of the stabilizing solution in the OMNIgene kit, viral culture was not performed on saliva (2). Viral culture was performed by serially diluting 100 µL of specimen 1:10 across the row of a 96-well plate in serum-free DMEM supplemented with 2x penicillin-streptomycin and 2x amphotericin B (Sigma-Aldrich, St. Louis, Missouri, USA). Vero CCL-81 cells were trypsinized and resuspended in DMEM + 10% FBS + 2x penicillin-streptomycin + 2x amphotericin B at 2.5 x 10^5 cells per mL. A 100 µL cell suspension was added directly to the clinical specimen dilutions and mixed gently by pipetting. The inoculated cultures were incubated at 37°C with 5% CO_2 and humidification, and observed daily for cytopathic effect. When cytopathic effect was observed, presence of SARS-CoV-2 was confirmed by RT-PCR. Serum specimens were analyzed using a validated pan immunoglobulin (Ig) enzyme-linked immunosorbent assay (ELISA) against the prefusion-stabilized extracellular domain of the SARS-CoV-2 spike protein to detect IgM, IgG and IgA anti-SARS-CoV-2 antibodies (3). Specimens were considered reactive if titers were ≥100 and results were presented as continuous variables.

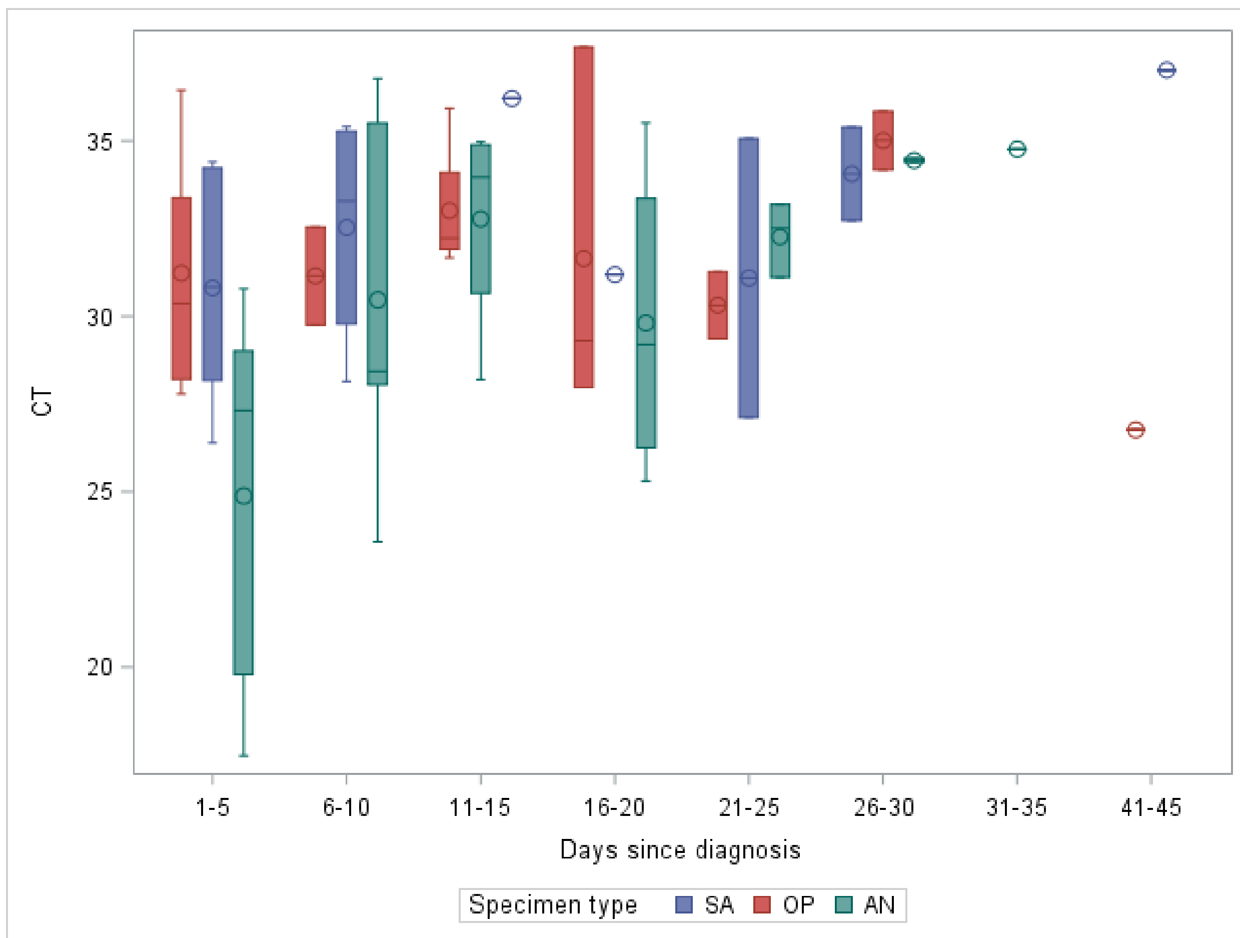
Surrogate neutralization antibodies were measured using Genscript's c-Pass surrogate neutralization kit (4). The results represent the percent that particular serum specimen blocked the interaction between the spike receptor binding domain (RBD) (virus) and Ace2 (cellular receptor). The higher the number, the more surrogate neutralization activity. We considered ≥ 70% as positive.

Supplemental Results

Ct value results

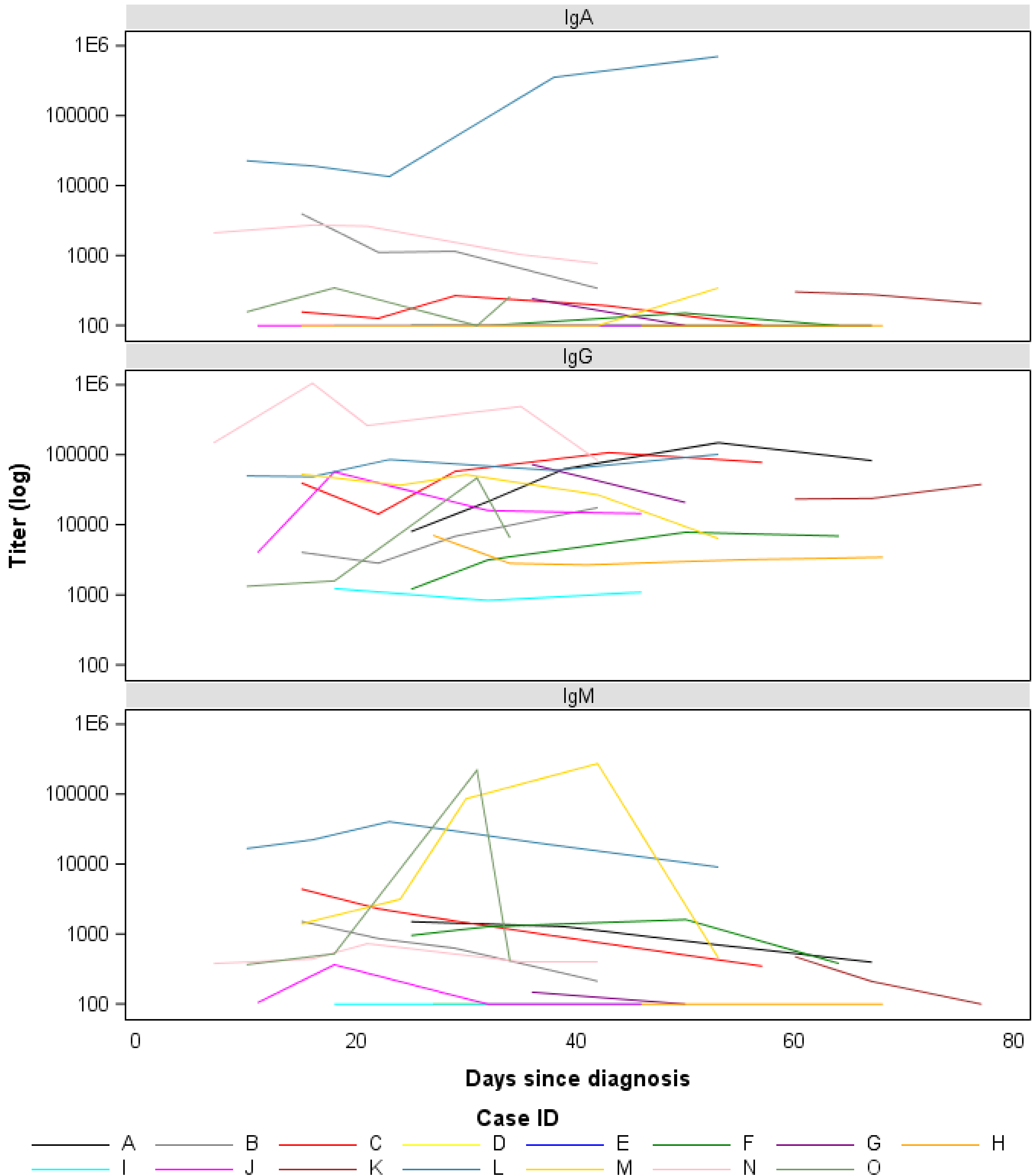
Median Ct values for AN, OP and SA specimens were 31.2 (IQR: 27.7 – 34.6), 31.7 (IQR: 29.3 – 34.2), and 33.5 (IQR: 29.5 – 35.3), respectively (Supplemental Figure 1).

Supplemental Figure 1. RT-PCR Cycle threshold (Ct) values per specimen type over time from end-stage renal disease (ESRD) participants on maintenance dialysis with SARS-CoV-2 infection (N=59 specimens)



SA, saliva; OP, oropharyngeal; AN, anterior nasal. The dot in the middle of the box indicates the mean, the horizontal line in the box indicates the median, the box indicates the interquartile range, the lines above and below the box indicate the range.

Supplemental Figure 2. Detection of SARS-CoV-2 IgM, IgG and IgA antibody titers among participants with positive antibodies (N=13) since diagnosis



Antibody to the SARS-CoV-2 spike protein was positive if equal to or greater than 100 (1E6=1000000). All antibody results are displayed in this figure, including those that are not considered positive (i.e. <100). Day of diagnosis is displayed as day 0 in the x-axis and represents the day of diagnosis recorded in the facility's medical record (either RT-PCR or antigen test).

References

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