

Expected Performance of Controls Included in the CDC 2019-nCoV Real-Time RT-PCR Diagnostic Panel

| Control Type | External Control Name | Used to Monitor | 2019 nCoV_N1 | -2019 nCoV_N2 | RP | Expected Ct Values |
|--------------|-----------------------|--------------------------------------------------------------------------------------|--------------|---------------|----|--------------------|
| Positive | nCoVPC | Substantial reagent failure including primer and probe integrity | + | + | + | < 40.00 Ct |
| Negative | NTC | Reagent and/or environmental contamination | - | - | - | None detected |
| Extraction | HSC | Failure in lysis and extraction procedure, potential contamination during extraction | - | - | + | < 40.00 Ct |

If any of the above controls do not exhibit the expected performance as described, the assay may have been set up and/or executed improperly, or reagent or equipment malfunction could have occurred. Invalidate the run and re-test.

RNase P (Extraction Control)

- All clinical samples should exhibit fluorescence growth curves in the RNase P reaction that cross the threshold line within 40.00 cycles (< 40.00 Ct), thus indicating the presence of the human RNase P gene. Failure to detect RNase P in any clinical specimens may indicate:
 - Improper extraction of nucleic acid from clinical materials resulting in loss of RNA and/or RNA degradation.
 - Absence of sufficient human cellular material due to poor collection or loss of specimen integrity.
 - Improper assay set up and execution.
 - Reagent or equipment malfunction.
- If the RP assay does not produce a positive result for human clinical specimens, interpret as follows:
 - If the 2019-nCoV N1 and N2 are positive even in the absence of a positive RP, the result should be considered valid. It is possible, that some samples may fail to exhibit RNase P growth curves due to low cell numbers in the original clinical sample. A negative RP signal does not preclude the presence of 2019-nCoV virus RNA in a clinical specimen.
 - If all 2019-nCoV markers AND RNase P are negative for the specimen, the result should be considered invalid for the specimen. If residual specimen is available, repeat the extraction