Technical Alert

Rapid SARS-CoV-2 (COVID-19) Combo NAAT Have Limitations

TO: Medical Staff, Clients, and Medical Laboratories

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SUBJECT: Rapid NAAT Combo tests (e.g., SARS / Flu A/B / RSV) limit the data that can be audited

Many companies have transitioned to SARS-COV-2 molecular tests that are combined with tests for other respiratory viruses such as influenza A and B and Respiratory Syncytial Virus (RSV). Like the COVID-only tests, these are approved by the FDA under Emergency Use Authorization (EUA). Some of these are Clinical Laboratory Improvement Amendment (CLIA) waived, which means they tends to mask the data that the instrument uses to make the result determination. Tests at DLS include Roche Liat and what we thought would be a transition to GeneXpert 4-Plex because Cepheid had stopped shipping COVID-only tests (Cepheid has since reinstated the COVID-Only test).

Unfortunately, the instruments do not always make the correct interpretation of the data due to software flaws or assumptions. DLS scientists recognized this very early (Possible False Positive SARS-CoV-2 (COVID-19) PCR (dlslab.com), Possible False Positive SARS-CoV-2 (COVID-19) NAAT (dlslab.com), and consequently implemented mandatory audits of any raw data available on all system before results are released. The criteria were:

Any test at Central with a cycle threshold (Ct) >27 is repeated. Any curve that shows inefficient amplification is questioned. For assays that have a minimum of 2 target sequences, any single target "hit" and/or Ct of >40 were resulted subject to additional testing on an alternate platform.

Although Combo assays seem to have maintained 2 target sequences for SARS-CoV-2, they now signal in the same fluorescent channel. Consequently, we cannot differentiate targets because fluorescence is generated by multiple reactions on the same channel, so we are uncertain what curves and/or Cts should be.

While the prevalence of COVID-19 in the community is low, the predictive value for a negative is excellent. In order to insure against potential combo false positives, DLS is implementing no-cost reflexing to another testing platform whenever the result is positive. We expect Lait positives at low Ct will correlate well with supplemental testing: however, we anticipate discrepants at higher Cts.

Because the multiplex reactions compete for the same reagents, presence of one virus may mask the presence of a co-infection with another virus. Whenever a specimen is positive for one virus (e.g., SARS-CoV-2), the negative results for other viruses (e.g., influenza A & B, RSV) are confirmed on another platform at no additional cost.

Results should be used in conjunction with other available information such as other lab data, clinical history, exposure risks, etc., and are not intended to be used as the sole means for diagnostic or patient management decisions.

If you have any questions, please call Dr. Amy Woron at 441-5436 or DLS client services at 589-5101.