PCR Testing: Advantages, Limitations and Interpreting Results

Advantages of PCR Testing

- Valuable for detecting specific pathogens that are difficult to culture in vitro or require a long cultivation period
- Significantly more rapid in providing results compared to culturing
    - Enables earlier informed decision making
    - Rapid diagnosis of bacteremia, particularly for low levels of bacteria in specimens
- Useful in detecting cases in extra pulmonary specimens which may be missed by smear and/or culture
- Valuable screening tool
    - PCR is still considered an adjunct test for certain diagnostic tests that still rely on smear and culture, such as tuberculosis

Limitations of PCR Testing

- PCR testing alone may be limited as a diagnostic tool
    - Still need culture for testing for drug/antibiotic susceptibility and genetic typing
- Post treatment diagnosis may be challenging
    - PCR detects dead organisms that may be shed for weeks after the patient stops showing symptoms. Unclear regarding persistence of infection. Detecting dead organisms at this stage may have no clinical relevance
- PCR results should not be used as the sole basis of a patient treatment management decision. All results should be interpreted by a trained professional in conjunction with review of the patient's history and clinical signs and symptoms
- <u>False positives and false negative results</u>
    - False negative results can arise from:
        - Improper sample collection/transport
        - Insufficient amount of specimen
        - Degradation of nucleic acids (typically RNA) during shipping or storage
        - Specimen collected prior to onset of symptoms or late in illness
        - Quantity of organisms is below detection limit
        - Non-homogeneous distribution of the organism of interest
        - Presence of amplification inhibitors in the specimen
        - Laboratory processing/testing errors
    - False positive results can arise from:

- Detecting contaminants introduced during specimen collection, transport or processing
- Detecting organisms representative of normal flora near specimen collection site, acid fast bacilli in water and contaminants in lab
- Mislabeling
- Specimen mix-up

Interpreting Results

- Negative result
  - A negative result means that there is no evidence of DNA or RNA of the target organism in the specimen tested. If no other etiology is identified and a specific infection is still clinically suspected, additional specimens should be collected and tested

- Positive result
  - A positive result indicates detection of DNA or RNA and confirms infection, but does not necessarily mean viable organism of interest is present or that the patient is contagious

Reasons for Discrepant Results Between PCR and Culture

PCR Positive and Culture Negative

- PCR is generally more sensitive than culture for detecting organisms of interest
- Subclinical colonization
- PCR detects non-viable organisms
- Administration of antibiotics; injured organisms
- False positive PCR results

PCR Negative and Culture Positive

- Presence of compounds in specimen that inhibit amplification of nucleic acids
- Laboratory testing error
- False negative PCR result

Suggest repeat testing and/or testing with antigen detection methods and/or other well-established DNA-amplification test. Please contact Monterey County Public Health Laboratory for more information.

References

Guidance for Clinicians on the Use of RT-PCR and Other Molecular Assays for Diagnosis of Influenza Virus Infection. http://www.cdc.gov/flu/professionals/diagnosis/molecular-assays.htm

Yang, S. and R.E. Rothman. 2004. PCR-based diagnostics for infectious diseases: uses, limitations, and future applications in acute-care settings. https://www.academia.edu/3577385/PCR-based_Diagnostic_for_Infectious_Diseases