UCLA Health Internal Guidelines for the Use of Plasma Cell-free DNA Metagenomic Nextgeneration Sequencing Assay (mNGS) (performed by Karius Inc. or UCSF)

Scope: These guidelines apply to the clinical use of plasma mNGS (performed by Karius Inc. or UCSF) for UCLA Health patients, both pediatric and adult. The ordering of plasma mNGS is restricted to Infectious Diseases providers at UCLA. This document is intended to provide guidance for all providers considering the use of plasma mNGS for their patients.

Background: Plasma mNGS assays have the ability to indiscriminately sequence millions of cfDNA fragments in a parallel high-throughput fashion with subsequent processing through a bioinformatics database, which in theory holds the promise of detection of various pathogens from single plasma sample. Real-world data of clinical impact of plasma mNGS testing are emerging (1-9). UCLA Health investigators have also published the clinical impact of this testing in our patient population (10). These guidelines are based on published data and are intended to optimize the use of plasma mNGS for patient care.

Known Limitations of Pathogen Detection by plasma mNGS assays.

- 1. Current plasma cf-mNGS assays do not detect RNA pathogens.
- 2. Sensitivity issues (false negatives): Plasma mNGS assays do not have the sensitivity of dedicated single microbiologic tests to detect specific targets and should not be relied upon as the sole modality to rule out an infectious etiology.
- 3. **Specificity issues (false positives)**: Plasma mNGS assays frequently detect DNA of viruses and commensal bacteria representing oral/respiratory or gut microbiome. Detection of these organisms is typically of little clinical impact and may lead to unnecessary testing or antibiotic therapy and prolonged hospital stay.
- 4. **Quantitative reporting (molecular per microliter, MPM)** generated by Karius Inc. has not been rigorously studied, may vary depending on the volume of blood or host factors at the time of collection, and can change drastically due to technical variations.
- 5. Antimicrobial resistance reporting by Karius is limited to selected pathogens and few select genomic markers of resistance (limited to mecA, mecC, vanA, vanB, KPC, and CTX-M) and does not encompass all potential resistance markers.

Recommendations for providers considering plasma mNGS testing for their patients.

Specific clinical syndromes where plasma mNGS testing has shown clinical utility
1. Culture-negative endocarditis or endovascular infections with blood cultures negative for 48-72 hours
2. Concern for zoonotic or vector-borne pathogens
3. Concern for certain fastidious pathogens (defined by ID)
Clinical scenarios where plasma mNGS testing may have clinical utility and may be
considered, but more research studies needed to clarify the clinical impact
1. Pneumonia in immunocompromised patients (does not replace bronchoscopy or
invasive tissue sampling given known sensitivity issue of plasma mNGS tests)
2. Invasive fungal infections
3. Complicated pneumonia in pediatric patients if no pathogen on respiratory specimen or pleural testing and if it will change management
4. Deep-seated abscess if aspiration from the source is not possible
Plasma mNGS testing should NOT be performed for the following indications
1. To perform serial molecular load monitoring; there are no published data validating the technology of quantification of organisms performed by Karius Inc. It is unknown whether the molecular load reported by Karius Inc (as MPM) can be reliably trended in different samples from the same patient.
2. To obtain molecular burden for known pathogen for reasons listed above
3. To confirm a known pathogen identified by other means
4. Repeat testing for same patient within a 2-month time period, with the exception of a new clinical illness that may warrant the use of the test.
5. Syndromes where plasma mNGS has been shown to have very low positive clinical impact:
a. Hospital-onset fevers
b. Bacterial pneumonia in adult patients
c. Meningoencephalitis, particularly among adult patients
d. Musculoskeletal infections
e. Neutropenic lever
1. Kule out infection in patients with suspected or established autoimmune diseases prior to starting immunosuppression

ID providers may consider ordering a virology hold for the purpose of mNGS assays when bronchoscopy or invasive tissue sampling has been done or another diagnostic test is pending, rather than sending the test up front.

References:

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