

## Identification of Microorganisms using Nucleic Acid Probes

Policy Number: AHS – M2097 – Identification of Microorganisms Using Nucleic Acid Probes	Prior Policy Name and Number, as applicable:
Effective Date: 08/01/2023	

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### I. Policy Description

Nucleic acid hybridization technologies utilize complementary properties of the DNA double-helix structures to anneal together DNA fragments from different sources. These techniques are utilized in polymerase chain reaction (PCR) and fluorescent resonance energy transfer (FRET) techniques to identify microorganisms (Khan, 2014).

### II. Related Policies

Policy Number	Policy Title
AHS-G2143	Lyme Disease
AHS-G2149	Pathogen Panel Testing
AHS-G2157	Diagnostic Testing of Common Sexually Transmitted Infections
AHS-G2158	Testing for Mosquito- or Tick-Related Infections
AHS-M2057	Diagnosis of Vaginitis Including Multi-Target PCR Testing

### III. Indications and/or Limitations of Coverage

Application of coverage criteria is dependent upon an individual’s benefit coverage at the time of the request.

- 1) The coverage status of nucleic acid identification using direct probe, amplified probe, or quantification for the microorganism's procedure codes is summarized in Table 1 below. "MCC" in the table below indicates that the test **MEETS COVERAGE CRITERIA**; while "DNMCC" tests indicates that the test **DOES NOT MEET COVERAGE CRITERIA**.

Microorganism	Direct Probe	Amplified Probe	Quantification
<i>Bartonella henselae</i> or <i>quintana</i>		87471 (MCC)	87472 (DNMCC)
<b>Candida species</b> (For vaginitis, please review AHS-M2057 Diagnosis of Vaginitis Including Multi-Target PCR Testing)	87480 (MCC) for vaginitis  87480 (DNMCC) for all other situations except vaginitis	87481 (DNMCC) for all situations	87482 (DNMCC) for all situations
<i>Chlamydia pneumoniae</i>	87485 (MCC)	87486 (MCC)	87487 (DNMCC)
<i>Clostridium difficile</i>	87493 (MCC)		
<i>Cytomegalovirus</i>	87495 (MCC)	87496 (MCC)	87497 (MCC)
<b>Enterococcus, Vancomycin-resistant (e.g., enterococcus vanA, vanB)</b>		87500 (MCC)	
<b>Enterovirus</b>		87498 (MCC)	
<b>Hepatitis B</b>		87516 (MCC)	87517 (MCC)
<b>Hepatitis G</b>	87525 (DNMCC)	87526 (DNMCC)	87527 (DNMCC)
<b>Herpes virus-6</b>	87531 (MCC)	87532 (DNMCC)	87533 (MCC)
<i>Legionella pneumophila</i>	87540 (MCC)	87541 (MCC)	87542 (DNMCC)
<b>Orthopoxvirus</b>		87593 (MCC)	
<i>Mycoplasma pneumoniae</i>	87580 (MCC)	87581 (MCC)	87582 (DNMCC)
<i>Mycoplasma genitalium</i>		87563 (MCC)	
<b>Respiratory syncytial virus</b>		87634 (MCC)	
<i>Staphylococcus aureus</i>		87640 (MCC)	
<i>Staphylococcus aureus, methicillin resistant</i>		87641 (MCC)	

- 2) Simultaneous ordering of any combination of direct probe, amplified probe, and/or quantification for the same organism in a single encounter **DOES NOT MEET COVERAGE CRITERIA**.

- 3) For any other microorganism without a specific CPT code, PCR testing **MEETS COVERAGE CRITERIA.**

### Policy Guidelines

A discussion of every infectious agent that might be detected with a probe technique is beyond the scope of this policy. Many probes have been combined into panels of tests. For the purposes of this policy, other than the respiratory virus panel, only individual probes are reviewed.

## IV. Table of Terminology

Term	Definition
AMA	American Medical Association
CDC	Centers of Disease Control and Prevention
CIDT	Culture-independent diagnostic test
CMV	Cytomegalovirus
CPT	Current procedural terminology
DFA	Direct fluorescent antibody testing
DNA	Deoxyribonucleic acid
EBV	Epstein Barr virus
EVD	Ebola virus disease
FDA	Food and Drug Administration
FRET	Fluorescent resonance energy transfer
H5N1	Hemagglutinin type 5 and neuraminidase type 1 (Avian Influenza A)
HDV	Hepatitis D virus
HEV	Hepatitis E virus
HIV 1	Human immunodeficiency virus type 1
HIV 2	Human immunodeficiency virus type 2
HPV	Human papillomavirus
HSV	Herpes simplex virus
HTLV-I	Human t lymphotropic virus type 1
HTLV-II	Human t lymphotropic virus type 2
IDSA	Infectious Diseases Society of America
ITS	Internal transcribed region
MRSA	Methicillin-Resistant Staphylococcus Aureus
NAATs	Nucleic acid amplification tests
NGU	Nongonococcal urethritis
PCR	Polymerase chain reaction
PID	Pelvic inflammatory disease
qPCR	Quantitative polymerase chain reaction
rDNA	Recombinant deoxyribonucleic acid
RNA	Ribonucleic acid
rRT-PCR	Real-time reverse transcriptase-polymerase chain reaction
RSV	Respiratory syncytial virus infection

Term	Definition
RT-PCR	Reverse transcriptase-polymerase chain reaction
SARS	Severe acute respiratory syndrome

## V. Scientific Background

Nucleic acid hybridization technologies, including polymerase chain reaction (PCR), ligase- or helicase-dependent amplification, and transcription-mediated amplification, are beneficial tools for pathogen detection in blood culture and other clinical specimens due to high specificity and sensitivity (Khan, 2014). The use of nucleic acid-based methods to detect bacterial pathogens in a clinical laboratory setting offers “increased sensitivity and specificity over traditional microbiological techniques” due to its specificity, sensitivity, reduction in time, and high-throughput capability; however, “contamination potential, lack of standardization or validation for some assays, complex interpretation of results, and increased cost are possible limitations of these tests” (Mothershed & Whitney, 2006).

## VI. Guidelines and Recommendations

### World Health Organization (WHO)

For detection of monkeypox, the WHO recommends “detection of viral DNA by polymerase chain reaction (PCR)” as the preferred laboratory test and recommends that any individual with a suspected case should be offered testing. They note that the best specimens for diagnosis are taken directly from the rash. Antigen and antibody detection may not be able to distinguish between orthopoxviruses (WHO, 2022).

### 2018 Infectious Diseases Society of America (IDSA)

Specific guidelines for testing of many organisms listed within the policy coverage criteria is found in the updated 2018 Infectious Diseases Society of America (IDSA) guidelines and recommendations titled, “A Guide to Utilization of the Microbiology Laboratory for Diagnosis of Infectious Diseases: 2018 Update by the Infectious Diseases Society of America and the American Society for Microbiology” (Miller et al., 2018). “This document is organized by body system, although many organisms are capable of causing disease in >1 body system. There may be a redundant mention of some organisms because of their propensity to infect multiple sites. One of the unique features of this document is its ability to assist clinicians who have specific suspicions regarding possible etiologic agents causing a specific type of disease. When the term “clinician” is used throughout the document, it also includes other licensed, advanced practice providers. Another unique feature is that in most chapters, there are targeted recommendations and precautions regarding selecting and collecting specimens for analysis for a disease process. It is very easy to access critical information about a specific body site just by consulting the table of contents. Within each chapter, there is a table describing the specimen needs regarding a variety of etiologic agents that one may suspect as causing the illness. The test methods in the tables are listed in priority order according to the recommendations of the authors and reviewers” (Miller et al., 2018).

## Centers of Disease Control and Prevention (CDC)

### *Candida Auris (C. auris)*

The CDC writes that “Molecular methods based on sequencing the D1-D2 region of the 28S rDNA or the Internal Transcribed Region (ITS) of rDNA also can identify *C. auris*.” The CDC further notes that various PCR methods have been developed for identifying *C. auris* (CDC, 2020a).

### *Chlamydia Pneumoniae (C. pneumoniae)*

The CDC writes that RT-PCR is the “preferred” method of detecting an acute *C. pneumoniae* infection. The CDC further notes that a positive culture should be confirmed by a second test, such as PCR (CDC, 2021a).

### *Ebola*

The CDC states that for diagnosis of Ebola, “there must be a combination of symptoms suggestive of EVD AND a possible exposure to EVD within 21 days before the onset of symptoms.” The CDC notes that PCR is one of the most common diagnostic methods (CDC, 2019a).

### *Giardia*

The CDC states that microscopy with direct fluorescent antibody testing (DFA) is considered the test of choice for diagnosing giardiasis, but rapid immunochromatographic cartridge assays, enzyme immunoassay kits, microscopy with trichrome staining, and molecular assays may be alternatively used as well. To obtain more accurate test results, the CDC recommends collecting three stool specimens from patients over the course of a few days. But, only molecular testing (e.g., DNA sequencing) can identify *Giardia* strains (CDC, 2021c).

### *Monkeypox Virus*

The CDC defines a suspect case of monkeypox as a “new characteristic rash, or meets one of the epidemiologic criteria and has a high clinical suspicion for monkeypox.” A probable case is defined as “no suspicion of other recent Orthopoxvirus exposure (e.g., Vaccinia virus in ACAM2000 vaccination) AND demonstration of the presence of Orthopoxvirus DNA by polymerase chain reaction of a clinical specimen OR Orthopoxvirus using immunohistochemical or electron microscopy testing methods OR Demonstration of detectable levels of anti-orthopoxvirus IgM antibody during the period of 4 to 56 days after rash onset.” A confirmed case of monkeypox is defined as “demonstration of the presence of Monkeypox virus DNA by polymerase chain reaction testing or Next-Generation sequencing of a clinical specimen OR isolation of Monkeypox virus in culture from a clinical specimen” (CDC, 2022b).

### *MRSA*

The CDC remarks that nucleic acid amplification tests (NAATs, such as PCR) “can be used for direct detection of *mecA*, the most common gene mediating oxacillin resistance in

staphylococci,” but will not detect novel resistance mechanisms or uncommon phenotypes (CDC, 2019b).

### *Mycoplasma Genitalium*

The CDC writes that “Men with recurrent NGU [nongonococcal urethritis] should be tested for *M. genitalium* using an FDA-cleared NAAT. If resistance testing is available, it should be performed and the results used to guide therapy. Women with recurrent cervicitis should be tested for *M. genitalium*, and testing should be considered among women with PID [pelvic inflammatory disease]. Testing should be accompanied with resistance testing, if available. Screening of asymptomatic *M. genitalium* infection among women and men or extragenital testing for *M. genitalium* is not recommended. In clinical practice, if testing is unavailable, *M. genitalium* should be suspected in cases of persistent or recurrent urethritis or cervicitis and considered for PID” (CDC, 2021d).

### *Non-Polio Enterovirus*

The CDC remarks that their laboratories “routinely” perform qualitative testing for enteroviruses, parechoviruses, and uncommon picornaviruses (CDC, 2018).

### *Respiratory Syncytial Virus (RSV)*

The CDC writes that real-time reverse transcriptase-polymerase chain reaction (rRT-PCR) and antigen detection tests are the most commonly used diagnostic tests, and are effective in infants and young children. However, the highly sensitive rRT-PCR is recommended to be used when testing older children and adults with RSV (CDC, 2020c).

### *Salmonella*

The CDC writes that diagnosis requires detection of the *Salmonella* bacteria, be it through culture or a “culture-independent diagnostic test (CIDT)” (CDC, 2019c).

### *Miscellaneous*

The CDC does not mention the need to quantify [through PCR] *Bartonella*, *Legionella pneumophila*, or *Mycoplasma pneumoniae*. However, PCR can be performed for both *Legionella pneumophila* and *Mycoplasma pneumoniae* specimen (CDC, 2020b, 2021b, 2022a). No guidance was found on Hepatitis G.

### **Committee on Infectious Diseases, American Academy of Pediatrics, 31st Edition (2018-2021, Red Book)**

The Committee on Infectious Diseases released joint guidelines with the American Academy of Pediatrics. In it, they note that “the presumptive diagnosis of mucocutaneous candidiasis or thrush usually can be made clinically.” They also state that FISH probes may rapidly detect *Candida* species from positive blood culture samples, although PCR assays have also been developed for this purpose (Pediatrics, 2018).

### **European Centre for Disease Prevention and Control (ECDC)**

On May 23, 2022, the ECDC released a rapid risk assessment of the monkeypox multi-country outbreak. They recommend that patients with probable cases should be tested with a “monkeypox virus specific PCR or an orthopoxvirus specific PCR assay which is then confirmed through sequencing” (ECDC, 2022b).

On June 2, 2022, ECDC released interim advice on risk communication and community engagement during the 2022 monkeypox outbreak in Europe. This is a joint report with the WHO regional office for Europe. They recommend speaking to your doctor about getting tested for monkeypox if you develop a rash with a fever or feeling of discomfort or illness (ECDC, 2022a).

### **United Kingdom Health Security Agency (UKHSA)**

The UKHSA recommends that monkeypox is diagnosed with a “PCR test on a viral swab taken from one or more vesicles or ulcers” (UKHSA, 2022).

## **VII. Applicable State and Federal Regulations**

### **Food and Drug Administration (FDA)**

Many labs have developed specific tests that they must validate and perform in house. These laboratory-developed tests (LDTs) are regulated by the Centers for Medicare and Medicaid (CMS) as high-complexity tests under the Clinical Laboratory Improvement Amendments of 1988 (CLIA '88). LDTs are not approved or cleared by the U. S. Food and Drug Administration; however, FDA clearance or approval is not currently required for clinical use.

As of 08/02/2022, a list of current U.S. Food and Drug Administration (FDA, 2022) approved or cleared nucleic acid-based microbial tests is available at: <https://www.fda.gov/medical-devices/vitro-diagnostics/nucleic-acid-based-tests>.

## **VIII. Applicable CPT/HCPCS Procedure Codes**

Procedure codes appearing in medical policy documents are only included as a general reference. This list may not be all inclusive and is subject to updates. In addition, codes listed are not a guarantee of payment.

<b>CPT</b>	<b>Code Description</b>
87471	Infectious agent detection by nucleic acid (DNA or RNA); Bartonella henselae and Bartonella quintana, amplified probe technique
87472	Infectious agent detection by nucleic acid (DNA or RNA); Bartonella henselae and Bartonella quintana, quantification
87480	Infectious agent detection by nucleic acid (DNA or RNA); Candida species, direct probe technique
87481	Infectious agent detection by nucleic acid (DNA or RNA); Candida species, amplified probe technique
87482	Infectious agent detection by nucleic acid (DNA or RNA); Candida species, quantification
87485	Infectious agent detection by nucleic acid (DNA or RNA); Chlamydia pneumoniae, direct probe technique

CPT	Code Description
87486	Infectious agent detection by nucleic acid (DNA or RNA); Chlamydia pneumoniae, amplified probe technique
87487	Infectious agent detection by nucleic acid (DNA or RNA); Chlamydia pneumoniae, quantification
87493	Infectious agent detection by nucleic acid (DNA or RNA); Clostridium difficile, toxin gene(s), amplified probe technique
87495	Infectious agent detection by nucleic acid (DNA or RNA); cytomegalovirus, direct probe technique
87496	Infectious agent detection by nucleic acid (DNA or RNA); cytomegalovirus, amplified probe technique
87497	Infectious agent detection by nucleic acid (DNA or RNA); cytomegalovirus, quantification
87498	Infectious agent detection by nucleic acid (DNA or RNA); enterovirus, amplified probe technique, includes reverse transcription when performed
87500	Infectious agent detection by nucleic acid (DNA or RNA); vancomycin resistance (eg, enterococcus species van A, van B), amplified probe technique
87516	Infectious agent detection by nucleic acid (DNA or RNA); hepatitis B virus, amplified probe technique
87517	Infectious agent detection by nucleic acid (DNA or RNA); hepatitis B virus, quantification
87525	Infectious agent detection by nucleic acid (DNA or RNA); hepatitis G, direct probe technique
87526	Infectious agent detection by nucleic acid (DNA or RNA); hepatitis G, amplified probe technique
87527	Infectious agent detection by nucleic acid (DNA or RNA); hepatitis G, quantification
87531	Infectious agent detection by nucleic acid (DNA or RNA); Herpes virus-6, direct probe technique
87532	Infectious agent detection by nucleic acid (DNA or RNA); Herpes virus-6, amplified probe technique
87533	Infectious agent detection by nucleic acid (DNA or RNA); Herpes virus-6, quantification
87540	Infectious agent detection by nucleic acid (DNA or RNA); Legionella pneumophila, direct probe technique
87541	Infectious agent detection by nucleic acid (DNA or RNA); Legionella pneumophila, amplified probe technique
87542	Infectious agent detection by nucleic acid (DNA or RNA); Legionella pneumophila, quantification
87563	Infectious agent detection by nucleic acid (DNA or RNA); Mycoplasma genitalium, amplified probe technique
87580	Infectious agent detection by nucleic acid (DNA or RNA); Mycoplasma pneumoniae, direct probe technique
87581	Infectious agent detection by nucleic acid (DNA or RNA); Mycoplasma pneumoniae, amplified probe technique



CPT	Code Description
87582	Infectious agent detection by nucleic acid (DNA or RNA); Mycoplasma pneumoniae, quantification
87593	Infectious agent detection by nucleic acid (DNA or RNA); orthopoxvirus (eg, monkeypox virus, cowpox virus, vaccinia virus), amplified probe technique, each
87634	Infectious agent detection by nucleic acid (DNA or RNA); respiratory syncytial virus, amplified probe technique
87640	Infectious agent detection by nucleic acid (DNA or RNA); Staphylococcus aureus, amplified probe technique
87641	Infectious agent detection by nucleic acid (DNA or RNA); Staphylococcus aureus, methicillin resistant, amplified probe technique
87797	Infectious agent detection by nucleic acid (DNA or RNA), not otherwise specified; direct probe technique, each organism
87798	Infectious agent detection by nucleic acid (DNA or RNA), not otherwise specified; amplified probe technique, each organism
87799	Infectious agent detection by nucleic acid (DNA or RNA), not otherwise specified; quantification, each organism

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## IX. Evidence-based Scientific References

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## X. Revision History

Revision Date	Summary of Changes
01/01/2022	Initial Effective Date
04/12/2022	Updated background, guidelines, and evidence-based scientific references. Literature review did not necessitate any modifications to the coverage criteria.
09/14/2022	Updated background, guidelines, and evidence-based scientific references. Literature review did not necessitate any modification to the coverage criteria. Edits made for clarity:  Removed “*DNMCC= Does Not Meet Coverage Criteria; MCC = Meets coverage criteria.” from beneath the table in CC1, as the definition was redundant with what was already provided in CC1.

Revision Date	Summary of Changes
	Revised code disclaimer statement
04/04/2023	<p>Updated background, guidelines, and evidence-based scientific references. Literature review necessitated the following changes in coverage criteria:</p> <p>Addition of “Orthopoxvirus” to the table in CC1. Code 87593 added in the “Amplified Probe” column in its row as “MCC” to allow for coverage of the amplified probe test for orthopoxvirus (monkeypox is an orthopoxvirus)</p> <p>CC2 edited for clarity, now reads: “Simultaneous ordering of any combination of direct probe, amplified probe, and/or quantification for the same organism in a single encounter DOES NOT MEET COVERAGE CRITERIA.”</p> <p>CC3 edited to remove specific list of organisms, as it was not all inclusive. Now reads: “For any other microorganism without a specific CPT code, PCR testing MEETS COVERAGE CRITERIA.”</p> <p>Added CPT code 87593 (new orthopoxvirus CPT code effective 7/26/2022)</p> <p>Committee approved 4/4/2023</p>