MAYO CLINIC LABORATORIES

Test Definition: KPNRP

KPC (blaKPC) and NDM (blaNDM) in Gram-Negative Bacilli, Molecular Detection, PCR, Varies

Overview

Useful For
Assessing pure isolates of gram-negative bacilli for mechanism of carbapenem resistance

Special Instructions
- Infectious Specimen Shipping Guidelines

Method Name
Real-Time Polymerase Chain Reaction (PCR) Using LightCycler with Amplified Product Detection Using Fluorescent Resonance Energy Transfer (FRET) Hybridization Probes

NY State Available
Yes

Specimen

Specimen Type
Varies

Shipping Instructions
1. See Infectious Specimen Shipping Guidelines in Special Instructions for shipping information.
2. Place specimen in a large infectious container (T146) and label as an etiologic agent/infectious substance.

Necessary Information
Organism identification and specimen source are required.

Specimen Required
The high sensitivity of amplification by PCR requires the specimen to be processed in an environment in which contamination of the specimen by *Klebsiella pneumoniae* (KPC) or New Dehli metallo-beta-lactamase (NDM) DNA is unlikely.

Supplies: Infectious Container, Large (T146)
Collection Container/Tube: Slant
Specimen Volume: Isolate
Collection Instructions:
1. Isolate the bacteria.
2. Bacterial organism must be in pure culture, actively growing. **Do not submit mixed cultures.**

Forms

If not ordering electronically, complete, print, and send a Microbiology Test Request (T244) with the specimen.

Reject Due To

Other	Agar plate, mixed cultures

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Varies	Ambient (preferred)		
	Refrigerated		

Clinical & Interpretive

Clinical Information

Nonsusceptibility to carbapenems in gram-negative bacilli by means of the enzyme KPC (*Klebsiella pneumoniae* carbapenemase) or NDM (New Dehli metallo-beta-lactamase) is becoming more common. The genes *bla*KPC and *bla*NDM encode KPC and NDM enzyme production, respectively. In addition to KPC and NDM production, there are other mechanisms of resistance to carbapenems in gram-negative bacilli, including production of other carbapenemases, or plasmid-encoded AmpC, or extended beta-lactamase production combined with decreased membrane permeability. Detection of carbapenemases by the modified Hodge test may be subjective and is not rapid. Testing for the minimum inhibitory concentration (MIC) determines the level but not the mechanism of resistance. PCR is a sensitive, specific, and rapid means of detecting of a specific portion of the genes encoding KPC and NDM production.

Reference Values

Not applicable

Interpretation

This PCR detects and differentiates both *bla*KPC and *bla*NDM. A positive KPC (*Klebsiella pneumoniae* carbapenemase) PCR indicates that the isolate carries *bla*KPC. A positive NDM (New Dehli metallo-beta-lactamase) PCR indicates the isolate carries *bla*NDM.

A negative result indicates the absence of detectable *bla*KPC or *bla*NDM DNA; however, false-negative results may occur due to inhibition of PCR, sequence variability underlying primers and, or loss of a plasmid carrying *bla*KPC and *bla*NDM.

Cautions

This assay should be used for testing of isolates of gram-negative bacilli. Request KNSRP / KPC (*bla*KPC) and NDM (*bla*NDM) Surveillance PCR, if testing directly from rectal or perirectal swabs is desired.

Supportive Data

The assay was validated using 159 gram-negative bacillus isolates, including 135 carbapenemase-producers (105 *bla*NDM positive and 30 *bla*KPC positive). The assay had 100% sensitivity and specificity for isolate testing compared with reference methods, including the modified Hodge test, testing for *bla*KPC using KPC (*Klebsiella pneumoniae* carbapenemase) PCR and testing for *bla*NDM by NDM (New Dehli metallo-beta-lactamase) PCR at the Health Protection Agency (HPA), London, UK.

Clinical Reference

1. Cunningham SA, Noorie T, Meunier D, et al: Rapid and simultaneous detection of genes encoding *Klebsiella pneumoniae* carbapenemase (*bla*KPC) and New Delhi metallo-beta-lactamase (*bla*NDM) in Gram-Negative Bacilli. J Clin Microbiol 2013;51:66-69
2. Multiplex Real-Time PCR Detection of *Klebsiella pneumoniae* Carbapenemase (KPC) and New Delhi Metallo-beta-lactamase (NDM-1) genes. Centers for Disease Control and Prevention 2011 (unpublished)
3. CLSI Document M100-S23, Vol.33 No.1, 2013. CLSI, Wayne, PA
4. New Carbapenem-Resistant Enterobacteriaceae Warrant Additional Action by Healthcare Providers. Centers for Disease Control and Prevention Health Alert Network, February 14, 2013

Performance

Method Description

Isolates are lysed in buffer to release their DNA. This assay amplifies and detects a specific portion of the genes encoding the KPC (*Klebsiella pneumoniae* carbapenemase) and NDM (New Dehli metallo-beta-lactamase) enzymes. The LightCycler instrument amplifies and monitors target nucleic acid sequences by fluorescence during PCR cycling. This is an automated PCR system that can rapidly detect amplified product development through stringent air-controlled temperature cycling and capillary cuvettes. The detection of amplified products is based on the fluorescent-resonance energy transfer (FRET) principle. For FRET product detection, a hybridization probe with a donor fluorophore, fluorescein, on the 3' end is excited by an external light source, which emits light that is absorbed by a second hybridization probed with an acceptor fluorophore LC-Led 610 (*bla*KPC specific) and LC-red 670 (*bla*NDM specific), on the 5' end. The acceptor fluorophore then emits a light of a different wavelength that can be measured with a signal that is proportional to the amount of specific PCR product. The detection process is completed in less than 1 hour using a closed tube system.(Cunningham SA, Noorie T, Meunier D, et al: Rapid and simultaneous detection of genes encoding *Klebsiella pneumoniae* carbapenemase (*bla*KPC) and New Delhi metallo-beta-lactamase (*bla*NDM) in gram-negative bacilli. J Clin Microbiol 2013;51:66-69)

PDF Report

No

Day(s) Performed

Monday through Friday

Report Available

2 days

Specimen Retention Time
30 days

Performing Laboratory Location
Rochester

Fees & Codes

Fees
- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their Regional Manager. For assistance, contact Customer Service.

Test Classification
This test was developed, and its performance characteristics determined by Mayo Clinic in a manner consistent with CLIA requirements. This test has not been cleared or approved by the US Food and Drug Administration.

CPT Code Information
87150 x2

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
KPNRP	KPC and NDM PCR	85502-3

Result ID	Test Result Name	Result LOINC® Value
SRC53	Specimen source	31208-2
35168	KPC PCR	49617-4
35169	NDM PCR	73982-1