UnityPoint Health METHODIST MICROBIOLOGY LABORATORY	Page 1 of 4	Section: UPM MICRO	Policy #: 11.011
	Approved by: see signature block at end of document		Date: 2/5/18 Review by: 2/5/18
	Supersedes: Date Revised: 4/1/14, 3/11/16, 2/5/18		
	Primary Responsible Parties: Marsha Bishoff Secondary Responsible Parties: Audrey Davis		
	CAP Standard:		
SUBJECT:	**NITROCEFIN DISK (β-LACTAMASE)**		

PRINCIPLE

Remel Nitrocefin Disk is a reagent-impregnated disk recommended for the use in qualitative procedures for the rapid detection of β-lactamase production by bacteria, particularly *Neisseria gonorrhoeae*, *Haemophilus influenzae*, *Moraxella catarrhalis*, *Staphylococcus* spp, *Enterococcus* spp, and anaerobic bacteria.

Nitrocefin is the substrate utilized in this test. β-lactamase hydrolyzes the β-lactam ring of nitrocefin, producing cephalosporanic acid. A distinctive color change is associated with this reaction wherein the pale yellow nitrocefin compound is converted to a pink end product upon hydrolysis. Aerobic and anaerobic β-lactamase producing bacteria effect this color change. Organisms not producing β-lactamase do not alter the pale yellow color of nitrocefin within the time limits of the test.

CLINICAL SIGNIFICANCE

The β-lactamase enzyme is produced by various organisms and is a mechanism of their resistance to penicillins and cephalosporins. A number of similar enzymes have been found in a number of bacterial species with somewhat different substrate specificities. Some selectively hydrolyze penicillin class antibiotics and are described as cephalosporinases; still other enzymes hydrolyze both. Test methods used to detect β-lactamase include iodometric, acidimetric and chromogenic cephalosporin procedures.

POLICY SCOPE

The scope of this policy applies to all Laboratory staff that prepares or performs testing on laboratory specimens at UnityPoint Methodist.

SPECIMEN

Biosafety Level 2
Organisms isolated from any cultured specimen.

REAGENTS
Nitrocefin Disk. Stored at 2-8 C until ready to use.

INSTRUMENTATION/EQUIPMENT
Wooden applicator stick or inoculating loop
Clean glass slide

QUALITY CONTROL
Beta lactamase discs are quality controlled when by lot # or shipment. QC is performed and recorded prior to testing patient isolates.

Strain:		**Expected Result:**
Staphylococcus aureus	ATCC 29213	Pink
Neisseria gonorrhoeae	ATCC 43069	Colorless

PROCEDURE:
1. Using forceps, place disk onto a clean microscope slide or in an empty petri dish.
2. Moisten each disc with one drop of distilled water.
3. With a sterilized loop or applicator stick remove several well-isolated similar colonies and smear onto disc surface.
4. Incubate disk for 5 minutes at room temperature. Observe disc for color change.
5. A positive reaction will be a pink or red color change on the area where the colonies were applied. (Color change does not usually develop over the entire disc.)
6. A negative result will show no color change on the disc.

REPORTING RESULTS
Results are entered in the culture work up. Comments "Beta-lactamase negative (Ampicillin susceptible)" or "Beta-lactamase positive (Ampicillin resistant)" are added to the culture observations when reporting.

PROCEDURAL NOTES/PROBLEM-SOLVING TIPS
1. The efficacy of this test in predicting the b-lactam resistance of micro-organisms other than *N.gonorrhoeae*, *Haemophilus influenzae*, *Moraxella catarrhalis*, staphylococci, and certain anaerobic bacteria is unproven.
2. Resistance of b-lactam antibiotics has been on rare occasions reported in some of the above organisms without the production of b-lactamases. In these cases, resistance mechanisms, such as permeability barriers, have been postulated. Therefore, the b-lactamase test should be used as a rapid supplement and not a replacement for conventional susceptibility testing.

3. For some strains of staphylocci, particularly *S. epidermidis*, an inducible b-lactamase has been described that might result in a false-negative b-lactamase reaction with a strain which is resistant to penicillin or ampicillin.

REFERENCES

Remel Package Insert Revised July 2014

MMCI Laboratory is a CAP accredited facility, as of 7/1/11 the responsibility of new and/or substantially revised policies and procedures will be restricted the Laboratory Director whose name appears on the CLIA certificate, whose signature appears below. The biennial review will be completed by the Administrative Director.

POLICY CREATION :		
Author:	***Ellyn Penning***	***April 1, 2014***
Medical Director:	***Douglas McGrady, MD***	***April 22, 2014***

MEDICAL DIRECTOR		
DATE	NAME	SIGNATURE
February 12, 2017	Elizabeth A. Bauer-Marsh, M.D.	Elizabeth A. Bauer-Marsh MD
SECTION MEDICAL DIRECTOR		
September 4, 2015	Lori Racsa, DO	L Racsa D.O.

REVISION HISTORY			
Rev	**Description of Change**	**Author**	**Effective Date**
0	Initial Release, replaces Cefinase Beta lactamase procedure	E Penning	4/1/14
1	Corrected grammar and updated reference	T Nuese	3/11/16
2	General reformatting. Updated negative QC organism. Updated reporting for Sunquest.	A. Davis	2/1/2018

Reviewed by

Lead	Date	Coordinator/ Manager	Date	Medical Director	Date
E Penning	4/1/14	Theresa R King	4/16/14	D McCarthy MD	4/22/14
		Teresa Nuese	3/15/16	L Racca D.O.	4/1/16
Marsha Bishoff	2/1/18	Audrey Davis	2/1/18	L Racca D.O.	2/5/18