HS NITRITE REAGENT KIT
MD-101102

Intended Use
For the quantitative determination of nitrite in urine.
NOT FOR USE IN UNPROFESSIONAL SETTINGS.

Summary and Principle
Detecting nitrite in urine helps identify urinary tract infections (UTIs) caused by certain bacteria[1]. Nitrite examination in urine is based on activity of nitrate reductase that is present in most Gram-negative uropathogenic rods, such as E. coli (Griess's examination). Nitrate reductase is, however, lacking from Pseudomonas aeruginosa and Gram-positive uropathogens such as Enterococcus spp. and Staphylococcus spp., and will therefore not be detected whatever their urinary concentration[2]. The positive detection of the enzyme requires, in addition, ingestion of nitrate by the patient (vegetables), its excretion into urine and a sufficient incubation time in bladder for reduction to nitrite.

MSSION Nitrite Reagent Kit contains a benzyl substituted salt which reacts with nitrite in the specimen to yield a colored complex read at 540 - 550 nm. The color intensity is proportional to the concentration of nitrite present to provide quantitative results.

Reagents
R1 and R2 reagents are provided ready to use. No preparation is required.
Nitrite R1 reagent contains a buffer and Nitrite R2 Reagent contains a buffer and a proprietary diazonium salt.

Reagent Stability and Storage
Store MISSION HS Nitrite Reagents at 2-8°C. When stored as directed, the reagent is stable until the expiration date stated on the label. The HS Nitrite Reagents has been tested to reflect shipping conditions and is stable for the lifespan of the product.

Cautions
MISSION HS Nitrite Kit is For Laboratory Use Only. May be harmful if inhaled or swallowed. Avoid contact of specimens with skin and eyes. Should contact occur, wash affected area with plenty of water.
DO NOT PIPETTE SPECIMENS BY MOUTH.

Instrumentation
HS Nitrite Kit are for use on Mission Diagnostics Affirm C200 [1]and Beckman AU680 Analyzers. Refer to instrument procedure instructions in the instrument manual provided with the specific analyzer.

Specimen Collection and Handling
Proper collection and storage of urine are essential for accurate urine nitrite testing. Collection: use a clean, dry, sterile container (plastic or glass). Storage: Room temperature (20–25°C) stable for <2 hours. Refrigerated at 2–8°C samples are stable for up to 24 hours.
Handle all urine samples as if potentially infectious.

Quality Control
Standard practice for Quality Control should be applied to this procedure. Store and handle reagents properly before and during use. Every laboratory should establish its own test requirements using controls and calibrators. Mission Diagnostics provides Calibrators to meet your program needs and which conform to NLCP Guidelines[3]:

Cat. No. MD-101202.01 – Nitrite Calibrator 5 mg/L
Cat. No. MD-101202.02 – Nitrite Calibrator 20 mg/L

Typical Performance Characteristics
The following performance data was obtained using the Affirm C200 and Beckman AU680 Analyzers. Other instruments may yield different performance data.

Linearity
The following results were obtained on an Affirm C200 and Beckman AU680 Analyzers using the MISSION HS Nitrite Reagent Kit on samples containing 0.50, 1.0, 2.5, 5, 10, 20 mg/L nitrite. The table below includes mean, standard deviation (SD) and Coefficient of Variation (CV) for each value.

Mean (mg/L)	SD	CV%
0.4834	0.0068	1.4
0.9902	0.0036	0.4
2.4910	0.0082	0.3
4.9750	0.0066	0.1
10.1062	0.1657	1.6
19.8856	0.0443	0.2

Precision
Studies performed on Affirm C200 and Beckman AU680 Analyzers. The precision of the assay was evaluated following a modification of NCCLS protocol EPT-T2. The within-run precision data was obtained by running two samples in replicates of 20 on the same day. The run-to-run data was obtained by running two samples in replicates over a five-day period.

[1] Also known as Zybio EXC200 Analyzer

Distributed by: Mission Diagnostics LLC, 1 Burton Dr., Meridith, NH 03246
Manufactured by: Diamond Diagnostics Inc., 333 Fiske St., Holliston, MA 01746
Technical Assistance Call: 1.508.429.0450
Email: support@diamonddiagnostics.com

Within-Run			Run-to-Run		
Mean (mg/L)	SD	CV%	Mean (mg/L)	SD	CV%
0.6936	0.0036	0.5	22.1465	0.7506	3.4
22.00	0.12	0.6	105.1581	2.8351	2.7

Analytical Specificity
Cross contamination studies have not been performed on Affirm C200 and Beckman AU680 Analyzers. Certain reagent/ instrument combinations used in sequence with this assay may interfere with reagent performance and test results. The existence of, or effects of, any potential cross contamination issues are unknown.

Test Conditions
For the data presented in this insert, studies using this reagent were performed on Affirm C200 and Beckman AU680 Analyzers using the parameters listed below.

Limitations
A sample with a nitrite concentration exceeding the analytical measuring range should be diluted with deionized water and reanalyzed incorporating the dilution factor in the calculation of the value.

Calibration
Calibration material should be used to calibrate the procedure. The frequency of calibration using an automated system is dependent on the system and the parameters used. If control results are found to be out of range, the test may need to be re-calibrated. Under typical operating conditions manufacturer calibration stability studies have shown the calibration curve will be stable for at least 14 days.

Method Parameters

Analyzer Specific Settings

Method type:	Endpoint
Slope:	positive
Units	mg/L
DOM wavelength	546
SUB wavelength	800
Sample volume	16
R1 volume	100
R2 volume	100
Blank rxn read (cycles)	16 - 20
Sample rxn read (cycles)	30 - 34

Calibration Settings

Calibration type	Linear
Reagent Blank required	No
Calibrator 1	0
Calibrator 2	5
Calibrator 3	20

Analytical Measuring Range (AMR)

Range: (low)	0
Range: (high)	30

BIBLIOGRAPHY

1. Clinical evaluation of nitrite test for the detection of bacteriuria. PMID: 9488933
2. Reliability of dipstick assay in predicting urinary tract infection. PMID: 25949979
3. National Laboratory Certification Program. General Laboratory Inspection Checklist / Report. OMB No. 0930-0158. November, 2002.

Distributed by: Mission Diagnostics LLC, 1 Burton Dr., Meridith, NH 03246
Manufactured by: Diamond Diagnostics Inc., 333 Fiske St., Holliston, MA 01746
Technical Assistance Call: 1.508.429.0450
Email: support@diamonddiagnostics.com