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Area	Laboratory-Chemistry
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Nitrite in Urine (Multistix 10 SG) - Royal Oak

Document Type: Procedure

I. PURPOSE AND OBJECTIVE:

A. The nitrite test depends on the conversion of nitrate (derived from the diet) to nitrite by the action of certain gram-negative bacteria in urine. Most Enterobacteriaceae are nitrate-reducing organisms, however some bacteria (e.g. Enterococcus) are not. Other bacteria (e.g. streptococcus, staphylococcus) further reduce nitrate to ammonia, nitric/nitrous oxide and nitrogen and therefore will give a negative test in the presence of significant bacteriuria.

B. At the acid pH of the reagent area, urinary nitrite reacts with p-arsanilic to form a diazonium compound. This compound in turn couples with 1,2,3,4-tetrahydrobenzo(h)quinolin-3-ol to produce a pink color.

C. This document describes the steps for this procedure to assist technologists.

II. SPECIMEN COLLECTION AND HANDLING:

A. **Fresh**, well-mixed, uncentrifuged urine. The conversion to nitrite requires at least four hours of bacterial incubation with urine in the bladder. Thus, first morning specimens are optimal. It is especially important to analyze a **fresh** specimen promptly, as false positives may occur with poorly collected/stored specimens because of bacterial proliferation from contaminants.

B. It is recommended that testing be done within one hour after voiding. Otherwise, immediately refrigerate the specimen and return to room temperature before testing.

III. REAGENTS/SUPPLIES:

A. Bayer Multistix 10 SG (#2161)

B. 1.4% weight for weight (w/w) p-arsanilic acid

C. 1.3% w/w 1,2,3,4-tetrahydrobenzo(h)quinolin-3-ol

D. 10.8% w/w buffer

E. 86.5% w/w non-reactive ingredients

IV. QUALITY CONTROL (QC):

A. Both Normal and Abnormal Kova-Trols are run and results are reported:

1. at the beginning of each shift
2. whenever a new bottle of reagent strips is opened
3. whenever troubleshooting warrants it

V. PROCEDURE:

A. Briefly dip the test area of the strip in fresh, **well-mixed** uncentrifuged urine.

B. While removing the strip, run the edge against the rim of the urine container to remove excess urine. Hold the strip in a horizontal position to prevent mixing of chemicals from adjacent reagent areas and/or contaminating the hands with urine.

1. If reading visually, compare the **NITRITE** reagent area to the corresponding Color Chart on the bottle label at **60 seconds**. Hold strip the close to color blocks and match carefully. Pink spots or pink edges should be interpreted as positive. Any degree of **uniform pink color development** is interpreted as positive. Comparison of the reacted reagent area against a white background may aid in the detection of low levels of nitrite, which may otherwise be missed.
2. If reading instrumentally, follow directions given in the Advantus procedure.

VI. REPORTABLE RANGE:

The Multistix 10SG has a color comparison chart with **THREE** color blocks. One negative block and two examples of positives. A positive nitrite suggests the presence of 10^5 or more organisms per mL, but color development is not proportional to the number of bacteria present.

VII. REFERENCE RANGE:

Negative

VIII. SENSITIVITY:

0.06 – 0.1 mg/dL nitrite ion

IX. INTERPRETATION:

The proportion of positive nitrite tests in case of significant infection depends on how long the urine specimens were retained in the bladder prior to collection. Identification of known positive cases with the nitrite test ranges from 40% with little bladder incubation to 80% with a minimum of 4 hours of

bladder incubation.

X. LIMITATIONS/INTERFERING SUBSTANCES:

A. The test is specific for nitrite, and will not react with any other

B. A negative result does not in itself prove that there is no significant bacteriuria. See the table below from Ames, Factors Affecting Urine Chemistry Tests, Miles Inc. 1992:

Test for Nitrite	
Urine Constituents or Situations Affecting Nitrite Results	**Clinitek Advantus**
Pink spots or pink edges	If pink spots or pink edges are observed, do NOT interpret as a positive result. IF any degree of uniform pink color development is observed, interpret as a positive result, suggesting presence of 10^5 or more organisms/mL. Color development is not proportional to number of bacteria
Urinary tract infections caused by organisms not containing reductase to convert nitrate to nitrite.	Negative results
When urine has not been retained in bladder long enough (4 hours +) to reduce nitrate to nitrite	May yield negative results
Lack of dietary nitrate in presence of organisms.	Negative results
High specific gravity urines	Sensitivity is reduced
Ascorbic acid (≥25 mg/dl) in urines	May cause false negative results if urine contains nitrite ion in concentrations of ≤0.06 mg/dL.

XI. REFERENCES:

1. Multistix 10 SG, Miles, Inc. Diagnostic Division, Elkhart, IN 46515, rev. 04/99
2. Henry, J.B., Clinical Diagnosis and Management by Laboratory Methods, 20th edition, Philadelphia, W.B. Saunders Co., 2001 pg.385.
3. Hundley, J.M. and Fleming, J.K., Urine Analysis American Society of Clinical Pathologists Workshop, Dearborn, MI, 1991.
4. Ames, Factors Affecting Urine Chemistry Tests, Miles, Inc., 1992.

Approval Signatures

Step Description	Approver	Date
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