Wake Forest Baptist Medical Center	Routine Urinalysis – Clinitek Advantus CL – U02	Dept:	Clinical Core Lab-Urinalysis Section
		Effective Date:	June 29, 1993
		Revised Date:	November 13, 2019
		Contact:	Clinical Core Lab-Hematology Management
Name & Title: Gregory J. Pomper, MD Medical Director of Pathology Laboratories		Date:	11/22/19
Signature: GJP			

1) **General Procedure Statement:**
   a. **Scope:** To provide laboratory testing personnel with instructions for performing laboratory procedures as deemed appropriate by industry practices and regulatory agencies to assist in quality patient care.

   b. **Responsible Department/Party/Parties:**

i.	Procedure owner:	Clinical Core Laboratory Management-Hematology
ii.	Procedure:	Clinical Core Laboratory Personnel
iii.	Procedure prepared by:	Heather Lawson
iv.	Supervision:	Clinical Core Laboratory Management-Hematology Clinical Core Laboratory Specialist and Designees Medical Director Clinical Hematology
v.	Implementation:	Clinical Core Laboratory Management-Hematology Clinical Core Laboratory Specialist and Designees Medical Director Clinical Hematology

2) **Definitions:**
   N/A

3) **Procedure:**

PRINCIPLE

The Routine Urinalysis is a simple method used for detecting, and monitoring the course and treatment of various endocrine or metabolic abnormalities involving the Renal System. A Routine Urinalysis at WFBH includes the following constituents: color and appearance, pH, specific gravity, protein, glucose, ketones, bilirubin, blood, urobilinogen, nitrite, leukocytes and a microscopic exam of the urine sediment, if indicated or specifically ordered. The chemical constituents are derived using a Multistix 10 SG and a Clinitek Advantus strip reader. The Clinitek Advantus is a semi automated reflectance spectrophotometer. The instrument analyzes the color and intensity of the light reflected from the reagent area and converts the results into clinically meaningful units. No calculations are required.

SPECIMEN COLLECTION AND HANDLING

A properly labeled random fresh urine specimen (<2 hours at room temperature) collected in a clean, dry, covered container is the specimen of choice. All specimens should be free from fecal and vaginal contamination. Analysis should be done as soon as possible after arrival in the laboratory. Formed elements may disintegrate at varying rates depending on pH, osmolality and storage conditions. Any urine specimen

not examined within 1-2 hours of receipt in the laboratory should be refrigerated until analyzed. Refrigerated samples may be analyzed up to 24 hours after collection.
Ten (10) mL or more of urine is preferred for macroscopic and microscopic urinalysis. Allow refrigerated urine specimens to return to room temperature before testing.

NOTE: If a Urine Culture is ordered on the same specimen as a Urinalysis, the culture should be processed prior to performing the urinalysis to avoid contamination of the specimen.

Specimen Rejection Criteria

If a sample is received over 4 hours old from an in-patient location, discard the sample and notify the floor to collect a new specimen. If a sample is received over 4 hours old from an out-patient clinic, analyze the sample and enter the Smart Text .OLD under Urine Comment. Specimens that are visibly mucoid or bloody should not be analyzed on the Advantus. If an unacceptable specimen is received, call the ordering location and request a new, acceptable specimen from the patient. Credit the original order in the LIS with the reason for the rejection.

Important: Some urine specimens may have been collected during a critical procedure or by means of an invasive procedure; therefore, it is important to never dispose of an unacceptable specimen until the caregiver has been notified.

EQUIPMENT /REAGENTS

1. Bayer Clinitek Advantus
2. Olympus BX40 phase microscope
3. Centrifuge capable of 2,000 rpm
4. 12 ml Conical Centrifuge tubes
5. Glass slides and 22 X 22 coverslips
6. Disposable berol pipets
7. 16 x 100 Test Tubes
8. Refractometer and capillary pipets
9. Multistix 10 SG Dipsticks – Store reagent strips in the original bottle at room temperature (15-30^{0}C). Strips are stable in the original bottle protected from light until the expiration date on the bottle.
10. MAS UA Controls-Store at 2-8^0 C. Unopened vials are stable until the expiration date on the label. Once opened, vials are stable 6 weeks when stored tightly capped at room temperature (18-25^{0}C) or 3 months refrigerated (2-8^{0}C)

QUALITY CONTROL:

Two levels of controls are run on 1st shift each day to assure the Clinitek Advantus is operating within established laboratory and manufacturer specifications.

1. Allow the controls to come to room temperature.
2. At the **Ready/Run** screen on the instrument, select the **Menu** key then Select the **QC** key.
3. Enter the control ID- UA1.
    a. Select **A-Z** to enter alphabetic characters.
    b. Select **Enter** to return to the numeric keypad.
4. Press **Enter**.
5. The display will prompt you with "Place strip". Mix the control tube by inversion. Dip a Multistix reagent strip into the control solution. Immediately remove the strip running the edge of the strip against the control tube to remove excess liquid.

6. Place the reagent strip on the loading station platform with the reagent pads facing up.

7. Repeat steps 3-6 for control ID- UA2. The strips will automatically advance along the loading station platform, under the readheads to be processed and then into the waste bin.
8. After the control(s) have been processed, the values will automatically print. Attach the printout to the 1st shift Atlas control printout and file the results in the QC notebook.
    a. If the control values are not "in" expected range, reclean the fixed platform, taking care to carefully clean the calibration bars. Rerun the controls.
    b. If control values are still "out" of expected range, open new controls and rerun.
    c. If control values are not recovered, do not process patient samples. Notify supervisor.
    d. File both sets of values in the QC notebook. Record the problem and follow-up action(s) on the Action Log.
9. After all controls are run, select **Return to Ready/Run** to exit the QC screen.
10. Record the control results on the Urinalysis QC Log. Result the controls in the LIS through the Result Activity window

CALIBRATION-ADVANTUS

Calibration of the Clinitek Advantus is performed at each readhead immediately before each reagent strip is read. The fixed platform has two white calibration bars that are positioned under each readhead. As a strip comes into position under a readhead, the instrument reads the calibration bar and calibrates for that scanning cycle. The instrument then scans the reagent strip and stores the data in memory. To get a printout of the last successful calibration: **Ready/Run screen→ Menu→ Print→ Calibration confirmation**

CALIBRATION-REFRACTOMETER

Calibration or the Zero Setting of the refractometer is checked daily with DH_2O. If the reading is more than 0.5% or ½ a division from zero adjustments need to be made. To adjust, remove the cement seal from the calibration opening. Push a jeweler's screw driver through the cement seal into the calibration opening. Turn the screw driver clockwise to increase the reading or counterclockwise to decrease the reading. The final motion should be clockwise. Reseal the opening with caulking compound. Calibration/recalibration is successful when the DH_2O reads 1.000.

MAINTENANCE

Bi-Weekly

1. Clean the exterior using a damp cloth and a mild detergent.
2. Clean the display screen with a soft, nonabrasive cloth dampened with a mild glass cleaner.

Note: Do not spray glass cleaner directly on the screen or use Kimwipes which may scratch the screen.

3. Remove and clean the push bar, fixed platform, moving table, hold down plate and liner with warm water and a mild detergent. Refer to pg. 37- 40 in the Clinitek Advantus Operator's Guide.
4. When cleaning the platform, avoid wiping across the two white calibration bars. Wet a cotton tipped applicator stick with water to clean the calibration bars. (If bars have scratches, marks or discoloration the platform must be replaced.)
5. Rinse each piece thoroughly and dry with paper towel. Allow calibration bars to air dry.
6. Reassemble the analyzer components.
7. Initial the Urinalysis Instrumentation Maintenance Log sheet indicating that the maintenance was performed.

As Needed

1. Change the thermal printer paper.
2. Lubricate the Push Bar Slide and Shaft.

PROCEDURE: UAMR-LAB348, UA-LAB2738, UANM-LAB2739

1. Urine samples are received in the lab from Central Processing. 2-10 ml of urine in a properly barcoded conical tube can be used for analysis.
2. If the instrument is at the screen saver display, touch the screen to display the Ready/Run screen.
3. Select the **ID** key from the **Ready/Run** screen to enter the specimen's ID. The ID can be entered using the numeric keyboard or the handheld barcode reader.
4. Enter the color and clarity then press **Enter**. The push bar moves to the left so that a reagent strip can be placed on the loading station.
5. At this point, the specimen can be run or another ID, color and clarity can be entered. If another ID is entered without a strip being detected, the instrument will automatically create a loadlist.
6. Mix the sample by inversion. Completely immerse all reagent areas on the strip in the specimen. While removing the strip, slowly run the edge of the entire strip against the side of the tube. **Do not blot excess liquid against a paper towel.**
7. Place the strip, reagent pads up, onto the loading station, to the right of the small embossed arrow and against the rear wall of the platform.
8. The strip is detected immediately and the push bar moves the strip to the readhead area where it is processed. The instrument will continue to move strips until the final strip is processed and moved to the waste area.
9. Exam the printed results. Attach the results to the Manual Urinalysis Worksheet. Perform any backup/confirmatory testing listed under Abnormal Urinalysis Results.

   Note: In the event that the urine dipstick results are unattainable via the Clinitek Advantus method, a manual dipstick with visual interpretation may be performed. Technologists should refer to the manufacturer's package insert which is included at the end of this procedure.

Abnormal Urinalysis Results

Positive Bilirubin

1. Positive bilirubin results will be reported with PCI (Possible Color Interference) appended to the result.
2. Yellow and Clear urines run on the Clinitek Advantus will automatically change the small bilirubin result to NEG before reporting.

Abnormal Color due to the presence of drug(s) or interfering substance

1. To report questionable interfering substance for macroscopic results due to abnormal urine colors, under Urine Comment, enter the Smart Text .INTRF. It is not necessary to flag each individual constituent.
2. Perform and report a manual specific gravity on samples processed by the Clinitek Advantus.

Grossly Bloody Specimen

1. For a grossly bloody specimen, spin down the urine and perform the dipstick on the supernatant. Use the original color and appearance when resulting.
2. Remix the sample. Perform the microscopic on the unspun urine. Under the UR Microscopic Comment dropdown box, result with Unspun Microscopic.

Specific Gravity

1. On the Advantus , samples with specific gravities ≤ 1.005 or ≥ 1.030 are reported. In the event that a specific gravity result is questioned, due to abnormal color or otherwise, it may be confirmed on the TS meter (refractometer).

Perform a Refractometer specific gravity as follows:

a. Be sure that the cover and prism (reading surface) are clean. If not, clean the surfaces with a drop of distilled water and a lint free cloth. Allow to dry.
b. Close the cover. Holding the Refractometer horizontally, apply a drop of urine at the notched bottom of the cover so that it flows over the prism surface by capillary action.
c. Point the Refractometer toward a light source at an angle that gives optimal contrast.
d. Read directly from the specific gravity scale where the line intersects between the light and dark contrast.

Extended Urinalysis

On Extended Urinalysis (LAB2740) Perform an SSA and Polarize exam (See Extended Urinalysis Procedure). Samples which auto-file will prompt for the UR Polarized Exam and the Urine Protein SSA to be resulted. If samples do not auto-file these tests must be resulted before saving and final verifying the test.

MICROSCOPIC EXAM

If a Urinalysis/w Microscopic if indicated (LAB 2738) order does not need a microscopic exam, the Urine Comment will auto answer with NMN (Microscopic Not Necessary) and the results will Final Verify. For urines needing a microscopic, the Urine Comment will auto answer with MICROW (Microscopic Necessary). Negative urine microscopics (no significant findings) will auto answer UR Microscopic Comment with NMU (Negative Microscopic Urine).

A microscopic exam will not be performed:

a. Unless a UAMR or UAXT2 urinalysis is ordered.
b. If a urine is yellow and clear with all negative chemical reactions
c. If a urine is yellow and clear with only a positive Urobilinogen
d. If a urine is yellow and clear with only a positive Ketone ≤ 15
e. If a urine is yellow and clear with only a positive Glucose OR
f. If a urine is yellow and clear with only a positive Glucose and positive Ketone ≤ 15

Samples that do not need a microscopic exam according to the above criteria will auto-file and Final Verify.

Samples requiring a microscopic exam continue with #A below.

Procedure for Manual Microscopic Exam

A. Spin 2 -10 mls of urine in a conical tube for 5 minutes at 2,000 rpm. Urines with volumes <2 mls should be examined unspun.
B. Pour/aspirate off the supernatant and resuspend the sediment in approximately 1ml of urine. With a 10ml volume, this results in an approximate 10:1 sediment concentration. Microscopic results are based on a 10:1 sediment concentration.
C. Using a disposable pipet, place a drop of the sediment on a glass slide and coverslip.
D. Using the Phase Contrast Microscope, quantitate the cellular elements by averaging the findings of 10 high power fields. Casts are quantitated using 10 low power fields.
E. Microscopic constituents should be correlated to the dipstick results for all specimens, whether autofiled or reviewed. One would expect large blood to correlate with TNTC RBC. However, the age or alkalinity of the specimen and/or non-nitrite producing bacteria could cause correlation discrepancies. Repeated inconsistencies with correlation of macroscopic and microscopic findings in consecutive runs could indicate a need for instrument maintenance or recalibration. Record any changes and/or verifications on the printed report and Action Log.

REPORTING/ INTERPRETING RESULTS:

Microscopic constituents should be correlated to the dipstick results. However, the age or alkalinity of the specimen and/or non-nitrite producing bacteria could cause correlation discrepancies. Repeated inconsistencies with correlation of macroscopic and microscopic findings in consecutive runs could indicate a need to clean the readheads, fixed platform, moving table and/or the holddown plate. If needed, clean the instrument and repeat the run.

1. All manual urinalysis results are entered into LIS through the Result Entry Activity window.
2. From the Outstanding List, double click on the patient to be resulted. This opens the Result Entry Activity window. Click Edit.
3. Click in the Component Value result box. Enter your result. If any comments need to be amended to the result, click on the Comment Field (paper icon) to the right of the result. This opens the Smart text window. Use Smart text phrases preceded by a period (.) or free text your result/comment. Click Accept.
4. Urine macroscopic comments should be made under Urine Comments (ex. INTRF-Interfering substance). Urine microscopic comments should be made under UR Microscopic Comment (ex. NMU).
5. Click Save →Verify →Final Verify.

For Manual Microscopic Exam

a. WBC, RBC and Casts are reported as a numeric range. EX: 0-3, >50, TNTC.
b. Bacteria and epithelials are reported as follows:

SEDIMENT VALUES AT 10X CONCENTRATION

	NORM/RARE	FEW	MOD	MANY	TNTC
Bact/hpf	<5	5-20	20-50	>50	packed
Epi/hpf	not in every field(rare)	1-5(normal)	5-25	25-100	>100

c. If WBCs are clumped, result Smart Text .CLMP in the WBC Comment field.

d. To report yeast, crystals, casts, etc.:

Yeast, Crystals and Sperm

1. Report the specific type of crystal or the presence of yeast and/or sperm under their respective Component Parameter. Do not quantitate. Sperm are reported regardless of the sex of the patient.

   Reporting Sperm on Pediatric patients:

   1. If sperm are seen on a patient **≤ 16 years old**, it must be confirmed.
   2. Acquire a fresh aliquot of the original urine specimen.
   3. Repeat the manual microscopic on the fresh aliquot. If the manual microscopic is POS for sperm, have a second technologist confirm the results. If the sperm results are confirmed POS by the second technologist, report Sperm as Present. Call the caregiver and document as a critical value.
   4. If the repeat microscopic is NEG, do not report Sperm.
   5. If another fresh aliquot of the original urine sample cannot be obtained, have a second technologist confirm the POS results. If the results are confirmed by the second technologist, report sperm as Present and append "Unable to verify with repeat sample".
   6. Save all spun tubes in the Fluid Rack under the appropriate day.

2. Any crystal which is considered to be pathologic (cystine, tyrosine, etc.) must be verified by confirmatory testing and the pathology resident on call before resulting in LIS. Refer to the chart on the following page for confirmatory tests.

Casts

1. Quantitate (0-2/lpf, 3-5/lpf, etc.) and report the specific type of cast seen. Always scan for casts on low power along the edges of the coverslip.
2. Report hyaline casts under UR Hyaline Casts and all other casts under UR Pathological Casts.

Fat Globules

1. If fat globules are suspected, view the microscopic using polarized light.
2. If positive, report under UR Microscopic Comment component's Comment field, Smart Text .POLP (Polarized exam positive). Free text, Free fat globules.

Dysmorphic RBCs

If seen, Dysmorphic RBCs (RBCs with blebs) should be noted in the UR RBCs Comment field as "Dysmorphic RBCs Present".

DIPSTICK NORMAL RANGES:

1. Color and appearance:	yellow/clear		
2. Specific gravity:	1.005 - 1.025	7. Ketones:	Negative
3. pH:	4.6 – 8.0	8. Bilirubin:	Negative
4. Protein:	Negative	9. Urobilinogen:	0.2-1.0 Ehrlich Units
5. Glucose:	Negative	10. Nitrite:	Negative
6. Blood:	Negative	11. Leukocytes:	Negative

MICROSCOPIC NORMAL RANGES

RBC/hpf	WBC/hpf	Hyaline Casts/lpf	Bact/hpf	Epithelials/hpf
0-3	0-5	0-2	0-5(rare)	0-5 (few)

CRITICAL VALUES:

Certain test values are critical to the care of a patient and the physician, charge nurse or other health care provider should be notified when they are reported.

Urine Ketones

Age <12 months old - Ketone ≥ 80

Document critical values in the LIS using the Smart text .CTRB (Called To And Read Back by) in the result comment field, with the time and the full name of the person taking the report. The person called should be asked to read back the result and patient information for verification purposes.

OTHER TESTS

UA tests UGL-LAB2752, UHA-LAB2766, UKET-LAB403, UPH-LAB2761, UBLD-LAB2735, USG-LAB2736 may be ordered individually using the stated test codes. These may be run on the Clinitek Advantus.

A Microscopic Only-LAB809, can be ordered on a UA that resulted as NMN or on a UANM or as an individual order.

PROCEDURE LIMITATIONS:

Urine test strips can be compromised if exposed for an extended time to the moisture in the air. Keep the Multistix 10 SG bottle tightly capped. Substances that cause abnormal urine color, such as drugs containing azo dyes, nitrofurantoin and riboflavin, may affect the color development and readability of the reagent pads. Refer to Siemen Product inserts for individual parameter interferences.

NOTES:

Resending Records from the Advantus Memory:

Up to 500 patient records and 200 QC records are stored in memory. Records can be recalled to the printer or to the LIS.

Procedure for resending one or more records to the LIS:

1. Recall a group of results.
    a. At the **Ready/Run** screen, select **Menu** then **Memory.**
    b. Recall a group of records:
        - All patient results
        - All QC results
        - Last batch of patient results
        - Search for stored results

The number of records in memory displays next to the first 2 options. The last batch of patient result tests are those tests run between the last pause in testing and the latest test. If the latest test is a QC test, it is not recalled.

    c. Select your Recall group. If you select Search for Stored results, enter the patient ID otherwise ↵ to start the search.
    d. After the search is completed.
        - The earliest record of the select group displays. The date and time the record was stored displays along with the SEQ# and ID for the record. All results are then listed. Positive results are flagged with an asterisk (*) and edited results with an exclamation point (!).
        - Use the movement keys ↑, ↓, ↑10, ↓10 to locate the first record to review.
2. Select → **Resend or** **Reprint**
3. Select one of the following options:
    a. Send only this result - Will send the displayed record
    b. Send a group of results - Will send specified records (enter first and last records)
    c. Send all patient results – Will send all records that were recalled
4. Select ← **Previous Screen** to return to the first record.

4) Review/Revision/Implementation:

a. Review Cycle: 2 years
b. Office of Record: Department of Clinical Core Laboratory-Hematology
c. All new procedures and procedures that have major revisions must be signed by the Laboratory Director.
d. All reviewed procedures and procedures with minor revisions can be signed by the designated section medical director.

5) Related Procedures:

Automated Urinalysis- AUWi CL-U01
General Information CL-HG02
Quality Control/Quality Assurance CL-UG01
Extended Urinalysis CL-U03

6) References, National Professional Organizations, etc.:

Henry, John. Clinical Diagnosis and Management by Laboratory Methods. Twentieth Edition. Philadelphia: W.B. Saunders Company, 2001.

Multistix 10 SG Package Insert. Siemens Healthcare Diagnostics, Inc. Rev. 03/2010

Clinitek Advantus Analyzer Operator's Manual, Siemens Healthcare Diagnostics Inc. 2009.

Instructions for Use and Care of the AO® TS Meter (A Goldberg® Refractometer). AO American Optical Scientific Instrument Division. Buffalo, NY. 02/1977

7) **Attachments:**
Manual Urinalysis Worksheet CL-U02-1
Siemens Multistix SG 10 Package Insert.
Urinary Sediment; A Textbook Atlas, TABLE 5-1 Urinary Crystals

8) **Revision Dates:**

Review Date	Revision Date	Signature
	4/11/15	Edelina Oliphant
	4/28/17	Edelina Oliphant
	5/14/17	Edelina Oliphant
	11/30/17	Edelina Oliphant
	07/20/2018	Heather Lawson
	11/13/19	Heather Lawson
18 Nov 19		

TABLE 5-1.—Urinary Crystals

CRYSTAL	COLOR	SHAPE	PH OF URINE	SOLUBLE IN	INSOLUBLE IN
NORMAL CRYSTALS					
Uric acid	Yellow (most common), colorless, reddish brown	Rhombic, whetstone, spears, needles, barrels	Acid	NaOH, heat (slight)	HCl, CH_3COOH, alcohol
Amorphous urates	Yellowish red, pink (sediment)	Amorphous, granules	Acid, neutral	NaOH, heat	HCl, CH_3COOH
Calcium oxalate	Colorless	Octahedral, dumbbell, round	Acid, neutral, slightly alkaline	Dilute HCl, HNO_3, NaOH (slight), heat	CH_3COOH
Hippuric acid	Colorless, pale yellow	Needles, rhombic plates, six-sided prisms	Acid, neutral, slightly alkaline	NaOH, heat, ether, alcohol	CH_3COOH
Amorphous phosphate	Colorless, white (sediment)	Amorphous, granules	Neutral, alkaline	HCl, CH_3COOH	NaOH, heat
Triple phosphate	Colorless	Prisms (coffin lid), feathers	Neutral, alkaline	HCl, CH_3COOH	NaOH, heat
Calcium phosphate	Colorless	Flat irregular plates, wedge-shaped prisms, granules	Slightly acid, neutral, alkaline	HCl, CH_3COOH	NaOH, heat
Calcium carbonate	Colorless	Dumbbell, granules	Neutral, alkaline	HCl + $CO_2\uparrow$ CH_3COOH + $CO_2\uparrow$	NaOH, heat
Ammonium urate	Yellowish brown	Scorpion, thorn apple, spheres	Alkaline	NaOH + $NH_3\uparrow$ HCl, heat (slow)	
ABNORMAL CRYSTALS					
Tyrosine	Colorless, yellow	Fine silky needles	Acid	NaOH, HCl, heat	CH_3COOH, alcohol, ether
Leucine	Yellowish brown	Spheroids with central striations	Acid	NaOH, hot CH_3COOH, heat	HCl, room temperature CH_3COOH, ether
Cystine	Colorless	Hexagonal plates	Acid	NaOH, HCl, NH_4OH	CH_3COOH, alcohol, ether, boiling H_2O
Cholesterol	Colorless	Flat plates with corners chipped out	Acid, neutral	$CHCl_3$, ether, hot alcohol	H_2O, dilute acids, dilute alkalis
Sulfa	Colorless, yellowish brown, greenish brown, colored complex formed by Lignin test	Amorphous, fan-shaped, shocks of wheat	Acid	Strong CH_3COOH, NaOH, acetone	Dilute CH_3COOH
Bilirubin	Bile-stained	Granules, needles	Acid	CH_3COOH, HCl, NaOH, $CHCl_3$, acetone, ether	
Starch	Colorless, purplish blue-black with iodine, does not stain with Sudan III	Irregularly round with dark striations to the center, asymmetric "Maltese cross" in polarized light, may be confused with leucine, fat bodies			

Multistix® 10 SG • Multistix® 9 • Multistix® 9 SG • Multistix® 8 SG • Multistix® 7 • N-Multistix® SG • N-Multistix® • Multistix® SG • Multistix® • Bili-Labstix® Reagent Strips

Tests for Protein, Blood, Leukocytes, Nitrite, Glucose, Ketone (Acetoacetic Acid), pH, Specific Gravity, Bilirubin and Urobilinogen in Urine.

SUMMARY AND EXPLANATION / INTENDED USE: Bayer Reagent Strips for Urinalysis include test pads for protein, blood, leukocytes, nitrite, glucose, ketone (acetoacetic acid), pH, specific gravity, bilirubin and urobilinogen. *Please refer to the carton or bottle label to see which tests are included on the product you are using.*

Bayer Reagent Strips are for professional use in near-patient (point-of-care) and centralized laboratory locations. The strips are intended for use in at-risk patient groups to assist diagnosis in the following areas:[1-4]

- kidney function
- urinary tract infections
- carbohydrate metabolism (e.g., diabetes mellitus)
- liver function

The strips also measure physical characteristics, including acid-base balance and urine concentration. Test results can be used along with other diagnostic information to rule out certain disease states and to determine if microscopic analysis is needed.[1-4]

Bayer Reagent Strips are ready to use upon removal from the bottle and the entire reagent strip is disposable. The strips may be read visually requiring no additional laboratory equipment for testing. The strips can also be read instrumentally using the CLINITEK® family of Urine Chemistry Analyzers and the appropriate software; contact your product representative for further information.

Bayer Reagent Strips are for *in vitro* diagnostic use. They have been determined to be nonhazardous under the guidelines issued by OSHA in 29 CFR 1910.1200(d).

INFORMATION REGARDING CLIA WAIVER:

- The CLINITEK STATUS and CLINITEK 50 Analyzers are CLIA waived only when used with Bayer Reagent Strips, manufactured by Bayer HealthCare LLC.
- These tests are CLIA waived when read visually and when run on the CLINITEK STATUS and CLINITEK 50 Analyzers. A certificate of CLIA waiver is required to perform these tests in a waived setting. To obtain a Certificate of Waiver, contact your state department of health or visit the CMS web site for an application, Form CMS-116.
- Failure to adhere to the instructions for use, including instructions for limitations, intended use, and performing quality control testing, is off-label use, resulting in these tests being categorized as high complexity and subject to all CLIA regulations.

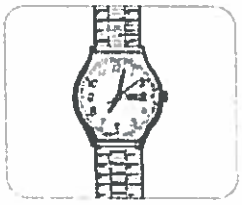
SPECIMEN COLLECTION AND PREPARATION: Collect freshly-voided urine in a clean container and test it as soon as possible. The container should allow for complete dipping of all reagent strip areas. A first-morning specimen is preferred but random collections are acceptable. Test the urine within two hours after voiding, sooner if testing for bilirubin or urobilinogen. If unable to test within the recommended time, refrigerate the specimen immediately and let it return to room temperature before testing. Work areas and specimen containers should always be free of detergents and other contaminating substances.[5]

Procedure

1. • **Collect** a fresh urine specimen in a clean, dry container
   • **Mix** well just before testing.
   • **Remove** one strip from the bottle.
   • **Replace** the cap.

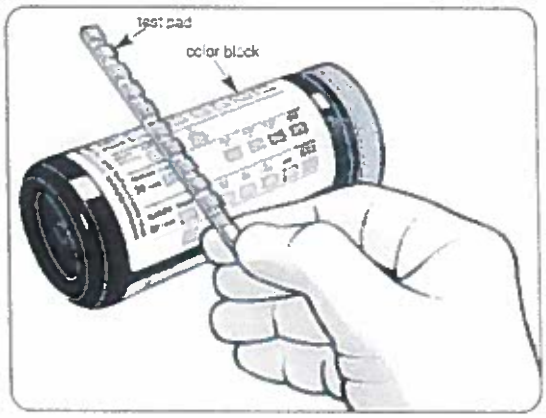
2. • **Dip** all the test pads of the strip into the urine
   • **Immediately remove** the strip

• If reading the strip visually, **start timing.**



3. **Drag** the *edge* of the strip against the container rim to remove excess urine.

4. a. **If reading visually.**
   • **Compare** each test pad to the corresponding row of color blocks on the bottle label.
   • **Read** each pad *at the time shown on the label*, starting with the shortest time.
   • **Hold** the strip close to the color blocks and match carefully.
   • **Read** the pads in good light.



b. **If using a CLINITEK instrument,** carefully follow the directions given in the appropriate instrument operating manual. The instrument will automatically read each test pad at a specified time.

5. Report the results to the lab supervisor or physician.

RESULTS: With visual use, results are obtained in clinically meaningful units directly from the Color Chart comparison. **With CLINITEK instruments,** the test pads are "read" by the instrument and the results are displayed or printed as soon as they are available.

QUALITY CONTROL: Test known negative and positive specimens or controls whenever a new bottle is first opened. Water should NOT be used as a negative control. Each laboratory should establish its own goals for adequate standards of performance. CHEK-STIX® Positive and Negative Control Strips provide a convenient basis for a quality control program.

STORAGE: All unused strips must remain in the original bottle. Transfer to any other container may cause reagent strips to deteriorate and become unreactive. Store at temperatures between 15°–30°C (59°–86°F). Do not use the strips after their expiration date. Do not store the bottle in direct sunlight and do not remove the desiccant from the bottle.

IMPORTANT NOTE: PROTECTION AGAINST EXPOSURE TO LIGHT, HEAT AND AMBIENT MOISTURE IS MANDATORY TO GUARD AGAINST ALTERED REAGENT REACTIVITY.

REAGENT PERFORMANCE:

Expected values for the "normal" healthy population and the abnormal population are listed below for each reagent.

Sensitivities listed for each reagent are the generally detectable levels of the analytes in contrived urines; however, because of the inherent variability of clinical urines, lesser concentrations may be detected under certain conditions. The percentage of clinical specimens correctly detected as positive increases with analyte concentration.

Performance characteristics are based on clinical and analytical studies and depend upon several factors: the variability of color perception; the presence or absence of inhibitory and matrix factors typically found in urine; and the laboratory conditions in which the product is used (e.g., lighting, temperature, and humidity). The strips should be read in good light, such as fluorescent; do not read in direct sunlight.

Each color block or instrumental result represents a range of values. Because of specimen and reading variability, specimens with analyte concentrations that fall between nominal levels may give results at either level. Results will usually be within one level of the true concentration. Exact agreement between visual results and instrumental results might not be found because of the inherent differences between the perception of the human eye and the optical systems of the instruments.

Limitations given for the reagents include specific substances and conditions that may affect the test results. **As with all laboratory tests, definitive diagnostic or therapeutic decisions should not be based on any single result or method.**

Substances that cause abnormal urine color may affect the readability of test pads on urinalysis reagent strips. These substances include visible levels of blood or bilirubin and drugs containing dyes (e.g., Pyridium, Azo Gantrisin, Azo Gantanol), nitrofurantoin (Macrodantin, Furadantin), or riboflavin. Levels of ascorbic acid normally found in urine do not interfere with these tests.

PROTEIN:

Expected values: Protein in urine can be the result of urological and nephrological disorders. In normal urine, less than 150 mg of total protein is excreted per day (24 hour period) (<15 mg/dL). Clinical proteinuria is indicated at greater than 500 mg of protein per day (strip result of >30 mg/dL). Positive results may also indicate tubular or overflow proteinuria in the absence of any glomerular abnormality or proteins of renal origin that may be excreted during infection. Urinary protein excretions can be temporarily elevated in the absence of renal abnormality by strenuous exercise, orthostatic proteinuria, dehydration, urinary tract infections, and acute illness with fever.[1,2] Clinical judgment is needed to evaluate the significance of Trace results.

Sensitivity: 15–30 mg/dL albumin.

Performance characteristics: The protein test pad is not specific for a particular protein, and proteins other than albumin can cause a positive response. The test is less sensitive to mucoproteins and globulins which are generally detected at levels of 60 mg/dL or higher.[3]

Limitations: A visibly bloody urine may cause falsely elevated results.[3]

BLOOD:

Expected values: Normally, no hemoglobin is detectable in urine (< 0.010 mg/dL or 3 RBC/µL). Occult blood occurs in urine as intact erythrocytes and hemoglobin, which can occur during urological, nephrological and bleeding disorders. Small amounts of blood (0.030–0.065 mg/dL or a strip result of Small) are sufficiently abnormal to require further investigation. The significance of the Trace reaction may vary among patients, and clinical judgment is required for assessment in an individual case. Blood is often, but not always, found in the urine of menstruating females.[1,4]

Sensitivity: 0.015–0.062 mg/dL hemoglobin.

Performance characteristics: The appearance of green spots on the reacted test pad indicates the presence of intact erythrocytes, while green color across the entire test pad indicates free hemoglobin. The test is equally sensitive to myoglobin as to hemoglobin. This test complements the microscopic examination; a hemoglobin concentration of 0.015–0.062 mg/dL is approximately equivalent to 5–20 intact red blood cells per microliter.[5]

Limitations: Captopril (captopril) may reduce the sensitivity. Certain oxidizing contaminants, such as hypochlorite, may produce false positive results. Microbial peroxidase associated with urinary tract infection may cause a false positive reaction.

LEUKOCYTES:

Expected values: Normal urine specimens generally yield negative results. An increase in leukocytes (≥10 leukocytes/µL) is an indication of pyuria and is found in nearly all diseases of the kidney and urinary tract; however, pyuria may often be present in non-infective conditions.[1] A strip result of Small or greater is a useful indicator of infection. Trace results may be of questionable clinical significance; however, Trace results observed repeatedly may be clinically significant.

Sensitivity: 5–15 white blood cells/hpf in clinical urine.

Performance characteristics: Leukocyte esterase is a reliable indicator of leukocytes in urine.[1] A positive reaction (Small or greater) at less than the 2 minute reading time may be regarded as a positive indication of leukocytes in urine.

Limitations: Elevated glucose concentrations (≥3 g/dL) may cause decreased test results. The presence of cephalexin (Keflex), cephalothin (Keflin), or high concentrations of oxalic acid may also cause decreased test results. Tetracycline may cause decreased reactivity, and high levels of the drug may cause a false negative reaction. Positive results may occasionally be due to contamination of the specimen by vaginal discharge.

NITRITE:

Expected values: Normally no nitrite is detectable in urine. Many enteric gram-negative organisms give positive results when their number is greater than 10^5/mL (0.075 mg/dL nitrite ion or greater).[2,3]

Sensitivity: 0.06–0.1 mg/dL nitrite ion.

Performance characteristics. The test is specific for nitrite and will not react with any other substance normally excreted in urine. Nitrite concentration during infection increases with the length of time the urine specimen is retained in the bladder prior to collection. A minimum of four hours of bladder incubation significantly increases the likelihood of obtaining a positive result.

Limitations: Pink spots or pink edges should not be interpreted as a positive result. A negative result does not rule out significant bacteriuria. False negative results may occur with shortened bladder incubation of the urine, absence of dietary nitrate, or the presence of nonreductive pathological microbes.

GLUCOSE:

Expected values: Small amounts of glucose (<15 mg/dL or 50 mg/day) are normally excreted by the kidney. These amounts are usually below the sensitivity level of this test but on occasion may produce a result between Negative and 100 mg/dL that is interpreted as a positive result. Results at the first positive level may be significantly abnormal if found consistently.[1,2]

Sensitivity: 75–125 mg/dL glucose.

Performance characteristics. The test is specific for glucose; no substance excreted in urine other than glucose is known to give a positive result. This test may be used to determine whether the reducing substance found in urine is glucose. If the color appears somewhat mottled at the higher glucose concentrations, match the darkest color to the color blocks.

Limitations: Ketone bodies reduce the sensitivity of the test; moderately high ketone levels (40 mg/dL) may cause false negatives for specimens containing small amounts of glucose (75–125 mg/dL) but the combination of such ketone levels and low glucose levels is metabolically improbable in screening.

KETONE:

Expected values: Normally, no ketone is detectable in urine (up to 2 mg/dL acetoacetic acid). In ketoacidosis, starvation or with other abnormalities of carbohydrate or lipid metabolism, ketones may appear in urine at levels of 10 mg/dL or higher before serum ketone levels are elevated. Clinical judgment is needed to determine the significance of Trace results, which may occur during physiological stress conditions such as fasting, pregnancy and frequent strenuous exercise.[1]

Sensitivity: 5–10 mg/dL acetoacetic acid.

Performance characteristics. The test reacts with acetoacetic acid in urine. It does not react with acetone or β-hydroxybutyric acid.

Limitations. False Trace results may occur with highly pigmented urine specimens or those containing large amounts of levodopa metabolites. Compounds such as mesna (2-mercaptoethane sulfonic acid) that contain sulfhydryl groups may cause false positive results or an atypical color reaction.

pH:

Expected values: The normal pH of urine can range from 4.6 to 8.0. Certain dietary conditions can produce acid or alkaline urines which can be useful in the treatment of some calculi.[1]

Performance characteristics: The pH test area measures pH values from 5–8.5 visually and 5–9 instrumentally, generally to within one unit of the expected result. pH readings are not affected by variations in the urinary buffer concentration.

Limitations: Bacterial growth by certain organisms in a specimen may cause a marked alkaline shift (pH > 8.0), usually because of urea conversion to ammonia.

SPECIFIC GRAVITY:

Expected values: The normal SG of urine ranges from 1.001–1.035. If the specific gravity of a random urine is 1.023 or greater, the concentrating ability of the kidneys can be considered normal.[1]

Performance characteristics: This test permits determination of urine specific gravity between 1.000 and 1.030, in general, it correlates within 0.005 with values obtained with the refractive index method. For increased accuracy, 0.005 may be added to readings from urines with pH >6.5. Strips read instrumentally are automatically adjusted for pH by the instrument. The Bayer SG test is not affected by the presence of radiopaque dyes as are the refractive index, urinometer, and osmolality methods.

Limitations: The Bayer SG test is dependent on ions in urine and results may differ from those obtained with other specific gravity methods when certain nonionic urine constituents, such as glucose, are present. Highly buffered alkaline urines may cause low readings, while the presence of moderate quantities of protein (100–750 mg/dL) may cause elevated readings.

BILIRUBIN:

Expected values: Normal adult urine contains about 0.02 mg/dL of bilirubin, which is not detectable by even the most sensitive methods. Even trace amounts of bilirubin are sufficiently abnormal to require further investigation.[1] Since very small amounts of bilirubin (0.1 mg/dL or greater) may be found in the earliest phases of liver disease, the user must consider whether the sensitivity of Bayer Reagent Strips to bilirubin is sufficient for the intended use. When very small amounts of bilirubin in urine are sought (e.g., in the earliest phase of viral hepatitis), ICTOTEST® Reagent Tablets should be the method of choice.

Sensitivity: 0.4–0.8 mg/dL bilirubin.

Performance characteristics: The test is specific for bilirubin and will not react with any other substance normally excreted in urine.

Limitations: Indican (indoxyl sulfate) can produce a yellow-orange to red color response that may interfere with the interpretation of a negative or positive reading. Metabolites of Lodine (etodolac) may cause false positive or atypical results. Atypical colors (colors that are unlike the negative or positive color blocks shown on the Color Chart) may indicate that bilirubin-derived bile pigments are present in the urine sample and may be masking the bilirubin reaction. These colors may indicate bile pigment abnormalities and the urine specimen should be tested further (e.g., ICTOTEST Reagent Tablets).

UROBILINOGEN:

Expected values: Urobilinogen is normally present in urine at concentrations up to 1.0 mg/dL (1 Ehrlich Unit/dL). A result of 2.0 mg/dL represents the transition from normal to abnormal and the patient and/or urine specimen should be evaluated further for hemolytic and hepatic disease. Evaluation of both the bilirubin and urobilinogen results helps in the differential diagnosis of jaundice as well as other liver and biliary disorders.[1]

Performance characteristics: This test area will detect urobilinogen in concentrations as low as 0.2 mg/dL (0.2 EU/dL) in urine. The absence of urobilinogen in the specimen cannot be determined.

Limitations: The test pad may react with interfering substances known to react with Ehrlich's reagent, such as p-aminosalicylic acid and sulfonamides. Atypical color reactions may be obtained in the presence of high concentrations of p-aminobenzoic acid. False negative results may be obtained if formalin is present. Strip reactivity increases with temperature; the optimum temperature is 22°–28°C (72°–79°F). The test is not a reliable method for the detection of porphobilinogen.

HELPFUL HINTS:

- Do not remove the strip from the bottle until immediately before it is to be used for testing. Replace the cap immediately and tightly after removing the reagent strip. Do not touch the test areas of the strip.
- Do not read any test pad after 2 minutes; color changes that occur after this time are of no diagnostic value.
- Discoloration or darkening of the test pads may indicate deterioration. If this is evident, or if test results are questionable or inconsistent with expected findings, the following steps are recommended: (1) confirm that the product is within the expiration date shown on the label; (2) check performance against known negative and positive control materials; (3) retest with fresh product. If proper results are not obtained, consult your local product representative, or contact the Customer Service Department, by calling 1-877-229-3711 (U.S. only), for advice on testing technique and results.
- Contamination of the urine specimen with skin cleansers containing chlorhexidine may affect protein (and to a lesser extent specific gravity and bilirubin) test results. The user should determine whether the use of such skin cleansers is warranted.
- It is especially important to use fresh urine to obtain optimal results with the tests for bilirubin and urobilinogen, as these compounds are very unstable when exposed to room temperature and light.

CHEMICAL PRINCIPLES OF PROCEDURES AND INGREDIENTS:
(based on dry weight at time of impregnation)

Protein. This test is based on the protein-error-of-indicators principle. At a constant pH, the development of any green color is due to the presence of protein. Colors range from yellow for "Negative" through yellow-green and green to green-blue for "Positive" reactions. **Ingredients:** 0.3% w/w tetrabromphenol blue; 97.3% w/w buffer; 2.4% w/w nonreactive ingredients.

Blood: This test is based on the peroxidase-like activity of hemoglobin, which catalyzes the reaction of diisopropylbenzene dihydroperoxide and 3,3',5,5'-tetramethylbenzidine. The resulting color ranges from orange through green; very high levels of blood may cause the color development to continue to blue. **Ingredients:** 6.8% w/w diisopropylbenzene dihydroperoxide; 4.0% w/w 3,3',5,5'-tetramethylbenzidine; 48.0% w/w buffer; 41.2% w/w nonreactive ingredients.

Leukocytes. Granulocytic leukocytes contain esterases that catalyze the hydrolysis of the derivatized pyrrole amino acid ester to liberate 3-hydroxy-5-phenyl pyrrole. This pyrrole then reacts with a diazonium salt to produce a purple product. **Ingredients:** 0.4% w/w derivatized pyrrole amino acid ester; 0.2% w/w diazonium salt; 40.9% w/w buffer; 58.5% w/w nonreactive ingredients.

Nitrite. This test depends upon the conversion of nitrate (derived from the diet) to nitrite by the action of Gram negative bacteria in the urine. At the acid pH of the reagent area, nitrite in the urine reacts with p-arsanilic acid to form a diazonium compound. This diazonium compound in turn couples with 1,2,3,4-tetrahydrobenzo(h)quinolin-3-ol to produce a pink color. **Ingredients:** 1.4% w/w p-arsanilic acid; 1.3% w/w 1,2,3,4-tetrahydrobenzo(h)-quinolin-3-ol; 10.8% w/w buffer; 86.5% w/w nonreactive ingredients.

Glucose: This test is based on a double sequential enzyme reaction. One enzyme, glucose oxidase, catalyzes the formation of gluconic acid and hydrogen peroxide from the oxidation of glucose. A second enzyme, peroxidase, catalyzes the reaction of hydrogen peroxide with a potassium iodide chromogen to oxidize the chromogen to colors ranging from green to brown. **Ingredients:** 2.2% w/w glucose oxidase (microbial, 1.3 IU); 1.0% w/w peroxidase (horseradish, 3300 IU); 8.1% w/w potassium iodide; 69.8% w/w buffer; 18.9% w/w nonreactive ingredients.

Ketone: This test is based on the development of colors ranging from buff-pink for a negative reading, to maroon when acetoacetic acid reacts with nitroprusside. **Ingredients:** 7.1% w/w sodium nitroprusside; 92.9% w/w buffer.

pH: This test is based on a double indicator principle that gives a broad range of colors covering the entire urinary pH range. Colors range from orange through yellow and green to blue. **Ingredients:** 0.2% w/w methyl red; 2.8% w/w bromthymol blue; 97.0% w/w nonreactive ingredients.

Specific Gravity: This test is based on the apparent pKa change of certain pretreated polyelectrolytes in relation to ionic concentration. In the presence of an indicator, colors range from deep blue-green in urine of low ionic concentration through green and yellow-green in urines of increasing ionic concentration. **Ingredients:** 2.8% w/w bromthymol blue; 68.8% w/w poly (methyl vinyl ether/maleic anhydride); 28.4% w/w sodium hydroxide.

Bilirubin: This test is based on the coupling of bilirubin with diazotized dichloroaniline in a strongly acid medium. The color ranges through various shades of tan. **Ingredients:** 0.4% w/w 2,4-dichloroaniline diazonium salt; 37.3% w/w buffer; 62.3% w/w nonreactive ingredients.

Urobilinogen: This test is based on the Ehrlich reaction in which p-diethylaminobenzaldehyde in conjunction with a color enhancer reacts with urobilinogen in a strongly acid medium to produce a pink-red color. **Ingredients:** 0.2% w/w p-diethylaminobenzaldehyde; 99.8% w/w nonreactive ingredients.

AVAILABILITY: Bayer Reagent Strips for Urinalysis are available in bottles of 100 strips: MULTISTIX® 10 SG (#2161); MULTISTIX® 9 (#2162); MULTISTIX® 9 SG (#2163); MULTISTIX® 8 SG (#2164); MULTISTIX® 7 (#2165); N-MULTISTIX® SG (#2176); MULTISTIX® SG (#2177); N-MULTISTIX® (#2178); MULTISTIX® (#2179); and BILI-LABSTIX® (#2180).

U.S. PATENT NUMBERS: Refer to the carton of the product you are using for applicable patent numbers.

TRADEMARKS:

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Bayer HealthCare

Bayer HealthCare LLC
Elkhart, IN 46514 USA