Blanchard Valley Health System: Point of Care Testing

Cardinal Health Urinalysis Analyzer Procedure

PRINCIPLE:

Cardinal Health Urinalysis Analyzer urine chemistry test system is intended for the in vitro qualitative and semi-quantitative measurement of the following parameters: blood, bilirubin, urobilionogen, ketones (acetoacetic acid), protein, nitrite, glucose, pH, SG (specific gravity), leukocytes, albumin and creatinine and the determination of the ACR (albumin: creatinine ratio). These measurements are useful in the evaluation of renal, urinary and metabolic disorders.

Blood: The test is based on pseudo-peroxidase reaction of hemoglobin. Oxygen is released, oxidizing tetramethylbenzidine, producing a color change from yellow through green to dark blue. The appearance of green spots on the reacted reagent area indicates the presence of intact RBC in the urine.

Bilirubin: This test is based on the coupling of bilirubin with diazotized dichloraniline in a strongly acid medium. The color range through various shades of pink to violet.

Urobilinogen: This test is based on the coupling of urobilinogen with diazotized p-methoxyaniline in a strongly acid medium. The colors range from beige through pink to dark pink.

Ketones: This test is based on the reaction of acetoacetic acid with nitroprusside, resulting in a color change from buff-pink to maroon.

Protein: This test is based on the color change of the indicator tetrabromphenol blue type in the presence of protein, producing a color change from yellow/green to blue.

Nitrite: This test is based on the conversion of nitrate to nitrite by the action of Gram negative bacteria in urine. At the acidic pH of the reagent area, nitrite in the urine reacts with sulfanilamide to form a diazonium compound. The diazonium compound couples with an aromatic compound to produce a pink color.

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 product colors ranging from blue through green to brown.

pH: This test is based on a double indicator (methyl red and bromothymol blue) principle that gives a broad range of colors, from orange, yellow, green, and blue.

Specific Gravity: This test is based on the pKa change of certain pretreated polymeric ion exchange resin in relation to ionic concentration. In the presence of an indicator, colors range from blue-green in urine of low ionic concentration through green and yellow-green in urines of increasing ionic concentration.

Leukocytes: This test is based on the color change ranging from beige to pink that occurs when esterase is hydrolyzed then coupled with diazonium salt to form a colored azo dye.

Albumin: This test is based on albumin binding to sulfonephthalein dye, producing a color ranging from pale green to aqua blue.

Creatinine: This test is based on the peroxidase-like activity of a copper creatinine complex that catalyzes the reaction of hydroperoxide and chromogen, producing color change from yellow through green to blue.

CLINICAL SIGNIFICANCE:

The intended use of the Cardinal Health™ Urinalysis Analyzer is to read the color change on the test pads found on the Cardinal Health™ urine test strips and to display and print the results. These measurements are useful in the evaluation of renal, urinary and metabolic disorders. . Microalbuminuria is an important adverse predictor of glycemic outcomes in pre-diabetes that may progress over a span of a number of years to overt nephropathy characterized by the presence of larger amounts of the protein albumin leaking through the kidneys' filter mechanism into the urine.

SPECIMEN:

A. COLLECTION AND PROCESSING:

Use only clean dry container to collect urine and test it as soon as possible. Do not centrifuge.

B. STORAGE AND PRESERVATION

If testing cannot be done within an hour after voiding, refrigerate the specimen immediately and let it return to room temperature before testing. The improperly stored (stored over 4 hours at room temperature, stored at high temperature (>30°C or >86°C) urine specimens may give inaccurate results.

REAGENTS, STANDARDS, AND CONTROLS:

Reagent	State	Storage	Expiration
Cardinal Health Urine Strips- UA10SGL	Unopened/Opened	Room Temperature and relative humidity 10-60%	Expiration Date on label
Cardinal Health Urine Strips- UA2ACR	Unopened/Opened	Room Temperature and relative humidity 10-60%	Expiration Date on label
Cardinal Health™ UA Control Dropper Bottle Set	Unopened	2-8°	Expiration Date on label
Cardinal Health™ UA Control Dropper Bottle Set	Opened	2-8°	After opening, the controls will remain stable until the expiration date stated on the label when stored at 2°-8°C between uses.

CONTROL PROCEDURE:

Test positive and negative quality controls (Cardinal Health UA Control Dropper Bottle Set) under the following conditions:

a. With each new shipment of reagent strips and every 30 days to check the storage of the reagent strips;
b. When using a new bottle of reagent strips;
c. When training instrument operators;
d. Whenever test results are in doubt;

Running Quality Control

1. Allow the Urine Control aliquot to reach room temperature (20-25°C) prior to testing. Perform the chemical test as for a patient specimen.
2. Verify that the lot number given on the value sheet enclosed in the package matches the lot number of the bottle of Urine Control. Promptly recap the bottle and return it to the refrigerator.
3. Gently swirl the control to assure good mixing, open the vial cap and apply Urine Control directly onto the reagent strips with a spraying technique. Hold the reagent strip horizontally, ensure good pad saturation and remove excess control by tilting the reagent strip on its edge on a paper towel. Each pad should be thoroughly moistened.
4. Read the urine reagent strips in accordance with the manufacturer's instructions as to timing and interpretation.
5. Record the results on the control chart provided.

6. Promptly recap the bottle and return the Urine Control to refrigerated storage.

If Quality Control falls outside of the stated values, the following sources of error may have occurred:

a. Improper technique or instrument setup. Check that reagent strip being used corresponds to the reagent strip name given on the status print out. Carefully repeat the QC procedure as described above.
b. Deterioration of the reagent strip test areas due to exposure to light, ambient moisture or heat. Obtain a fresh bottle of the Cardinal Health™ reagent strips being used and repeat the QC
c. Deterioration of the control solution. Obtain fresh control solution and repeat
d. Cardinal Health™ Urinalysis Analyzer instrument malfunction. Perform an initial instrument check. If the instrument/reagent strip performance check procedures cannot be successfully completed and an instrument malfunction or reagent strip problem is suspected, see Troubleshooting, or contact Technical Support for assistance.

EQUIPMENT:

A. Instruments:
Cardinal Health Urinalysis analyzer

B. Maintenance:
For routine maintenance, clean the strip holder daily or more often if sediment is seen, using soft gauze and distilled water. Avoid scratching the strip holder.

TEST PROCEDURE:

This procedure must be followed exactly to achieve reliable results.

1. Please refer to the box and bottle label for specific reagent areas on the product you are using. Confirm that the product is within the expiration date shown on the label.
2. Collect fresh, well-mixed urine specimen in a clean, dry container. Mix well immediately before testing.
3. Remove one strip from the bottle and replace the cap immediately.
4. Inspect the strip. If reagent areas are discolored, do not use the strip.
5. Read the strip on Cardinal Health Urinalysis Analyzer only: Touch the word “Measure” after power on.
6. Dip the test strip into the urine up to the last test pad for no more than 1 second.
7. Immediately wipe off excess urine on an absorbent paper. Lightly touch the edges of one side of the test strip on the absorbent paper.
8. Place the reagent strip onto the instrument’s strip holder without delay.
9. Touch the word “Start.”
10. The strip holder is automatically pulled into the instrument, where the strip is identified and read. Results are displayed or printed as soon as they are available.
11. Record the results you obtain, then discard the strip into a suitable trash container.

INTERPRETATION OF RESULTS:

Glucose Bilirubin, Ketone, Blood, Protein, Nitrite and Leukocytes are normally negative. Protein in trace amounts (10 mg/dL) is also considered normal. Urobilinogen is normal at < 2 mg/dL, (0.2-1.0 mg/dL). Specific Gravity normal range is 1.003 – 1.029. pH 5.0 – 8.0 can be considered to be within the reference range.

CALIBRATION:

A. Manual Calibration

Calibration procedure should be performed when:

- Changing product lot number
- Changing an item
- Test results are in doubt or different result is obtained than expected using quality control materials

CALIBRATION – Press “Calibration” icon at the main menu

Enter the lot number of the urine reagent strip to be measured. Perform the calibration.

Use the reagent strip dipped in distilled water

B. Automatic Calibration
The Cardinal Health™ Urinalysis Analyzer calibrates automatically before each measurement. The analyzer calibrates by reading the white check bar at the appropriate wavelengths to ensure accurate test results. The white check bar is tested on a reference spectrophotometer. The instrument performs a "self-test" and calibration each time it is turned on. Each time a test is run, the analyzer re-calibrates using a white check bar. Reflectance measurements from the bar must match the factory set calibration.

EXPECTED RESULTS:

Blood: Negative by result; 2~3 red cells per high power field are generally accepted as normal by microscopy.
Bilirubin: Detectable amounts of bilirubin are not normally present in urine. **Urobilinogen:** Male (0.3~2.1 mg/2 hours), Female (0.1~1.1 mg/2 hours). Results are sometimes expressed in Ehrlich units, 1 mg urobilinogen = 1 EU
Ketones: In starvation diets or in other instances of abnormal carbohydrate metabolism, ketones appear in the urine in excessively large amount before serum ketones are elevated.
Protein: Normally up to 20 mg/dL of protein in the urine is not considered pathological. **Nitrite:** Negative
Glucose: A small amounts of glucose (up to 30 mg/dL) may be present in normal urine. **pH:** 5~8, normal kidneys can produce urine with pH from 4.5~8.2, but with ordinary diet, urine pH is about 6.0.
Specific Gravity: 1.003~1.029, Adult on normal fluid intake: 1.016~1.022, Specific Gravity decreases with increasing age.
Leukocytes: Normal urine ordinarily yield negative results.
Albumin: : Albumin is normally present in urine at concentrations of less than 20 mg/L.14 Moderately increased albuminuria is defined as an albumin excretion rate of 30 ~ 299 mg/24 hours.15, 16 Urinary albumin excretions can be temporarily elevated by exercise, urinary tract infections, and acute illness with fever.
Creatinine: Creatinine is normally present in urine. There are no established reference values for creatinine in the urine however can be used to normalize other analytes found in a random urine sample at concentrations of 10 to 300 mg/dL (0.9 ~ 26.5 mmol/L).
Albumin to Creatinine Ratio: Albumin to Creatinine Ratio is normally at less than 30 mg albumin/g creatinine (3.4 mg albumin /mmol creatinine). Moderately increased albuminuria is indicated at a ratio result of 30 ~ 300 mg/g (3.4 ~ 33.9 mg/mmol) and severely increased albuminuria at a ratio result of > 300 mg/g (> 33.9 mg/mmol).

REPORTING RESULTS:

A. NORMAL VALUES:

Test	Normal Value
Leukocytes	Negative
Nitrate	Negative
Urobilinogen	<2.0
Protein	Negative
pH	5.0-8.0
Blood	Negative
Specific Gravity	1.003-1.029
Ketones	Negative
Bilirubin	Negative
Glucose	Negative
Albumin	NA
Creatinine	NA
Albumin/Creatinine Ratio	<30

LIMITATIONS AND INTERFERRING SUBSTANCES:

Blood: Elevated S.G or protein in urine may reduce the reactivity of the Blood test portion. Oxidizing contaminants, such as hypochlorite, may produce false-positive results. Microbial peroxidase associated with urinary tract infection may cause a false-positive result. Higher ascorbic acid concentrations (>50 mg/dL) may cause false-negative result at low level of blood.
Bilirubin: Metabolites of drugs, such as Pyridium and Serenium, which give a color at low pH may cause false-positive. Indican (indoxyl sulfate) can produce a yellow-orange to red color response which may interfere with the interpretation of negative or positive readings. Ascorbic acid (>25 mg/dL) may cause false-negative result.
Urobilinogen: The absence of urobilinogen in the specimen cannot be determined. The test area will react with substances known to derive Ehrlich's reaction, such as paraaminosalicylic acid. The test is not a reliable method for the detection of porphobilinogen.
Ketones: Highly pigmented urine or large amounts of levodopa metabolites containing urine may cause weak positive results. Some high S.G and low pH urine may give false-positive result. P.S.P. (phenolsulfonphthalein) may cause false-positive result.
Protein: Highly alkaline urine (>pH9) may cause false-positive result. Quinine, quinidine, chloroquine, trimethoprim, phenazopyridine, polyvinylprolidone (blood substituents) and the residues of disinfectants containing quaternary ammonium groups or chlorohexidine in the urinary vessel may cause false-positive.
Nitrite: Ascorbic acid (>25 mg/dL) may cause false-negative result with low level of nitrite containing (<0.03 mg/dL) urine. The negative result does not always mean that the patient is free from bacteriuria. Negative result may occur when urinary tract infections are caused by organism which do not contain nitrate reductase; when urine has not been retained in the bladder long enough (four hours or more) for reduction of nitrate to nitrite occur; or when dietary nitrate is absent.
Glucose: High S.G (>1.020) with high pH urine and ascorbic acid (>50 mg/dL) may cause false-negative result at the low level of glucose. Ketones reduce the sensitivity of the test. Moderately high ketone level (>40 mg/dL) may cause false-negative for specimens containing small amounts of glucose (<100 mg/dL). Reactivity may be influenced by urine S.G and temperature. If the color appears somewhat mottled at the higher glucose concentration, match the darkest color to the color block.
pH: If the excessive urine is remain on the strip because of improper test procedure, it is possible that the acidic buffer in protein portion comes out and affect the pH portion, then pH result may be decreased than the actual value. This phenomenon is called "run-over effect".
Specific Gravity: Highly buffered alkaline urine may cause diminished result, whereas highly buffered acidic urine may cause slightly elevated result.
Leukocytes: Large urinary protein excretion (>500 mg/dL) may cause false-negative result. Nitrofurantoin masks the reacted color to yellow. Tetracycline may cause false-negative result at a low level of Leukocytes. High concentration of glucose (>2000 mg/dL) may diminish this reaction at a low level of Leukocytes

Analyte	Concentration of Substance at which interference was observed	Change in Color Block output
Albumin	Calcium chloride ≥200 mg/dL, Fructose ≥80 mg/dL, Ascorbic acid ≥300 mg/dL, Citric acid ≥65 mg/dL, Sodium nitrite ≥8 mg/dL, Potassium chloride ≥1200 mg/dL, Sodium chloride ≥5000 mg/dL, Ribofavin ≥15 mg/dL, High specific gravity ≥1.050	-1
	Sodium bicarbonate ≥1350 mg/dL, Phenolphthalein ≥1050mg/dL, Theophylline ≥85 mg/dL, Sodium acetate ≥250 mg/dL, Acetaminophen ≥40 mg/dL, High pH ≥pH 9, Bilirubin ≥4 mg/dL, Hemoglobin ≥5 mg/dL, Blood ≥300 mg/dL	+1
Creatinine	Glycine ≥430 mg/dL, Sodium bicarbonate ≥1200 mg/ dL, Sodium-2-mercaptoethene ≥510 mg/dL, High pH ≥pH 9 -1	-1
	Calcium chloride ≥220 mg/dL, Sodium chloride ≥5200 mg/dL, Albumin ≥890 mg/dL, Theophylline ≥90 mg/dL +1	+1

PROCEDURAL NOTES:
This test is waived under CLIA ‘88 regulations. If a laboratory modifies the test system instructions, then the test is considered high complexity and subject to all CLIA requirements. A CLIA Certificate of Waiver is needed to perform CLIA waived testing.

REFERENCES:
Cardinal Health Urinalysis Analyzer Operator’s Manual, 2020-04
Cardinal Health Urine Strips UA10SGL Product Insert, 2020-04 Rev.0
Cardinal Health Urine Strips UA2ACR Product Insert, 2020-04 Rev.0

Laboratory Director Approval

Approved by _____ Date _____