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Document History:

Title: **Siemens Multistix 10 SG Reagent Strip and CLINITEK Novus 10 Urinalysis Cassette**

Site(s): **Shared Health**

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Approved by:	T Dembinski	**Date:**	16 May 2013

Approved by:	L Thorlacius	**Date:**	4 Mar 2021
Signature:	(Signature on File)		
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#	Details of Revisions:	Approval:	Date:
1	New document	T Dembinski	16-MAY-2013
2	Correction of SG upper ref range limit; standardization of procedural steps for consistency to related updated urinalysis SOPs.	H Malvern	13-MAR-2015
3	Amended reagent strip storage information under “Precautions for Handling Multistix…”(pg.3); updated references (pg.11)	T Dembinski	01-June-2017
4	Added new statement under Sample requirements “Grossly visibly bloody urines should be rejected” pg. 3.	T Dembinski	08-Sep-2017
5	Included specific details for Novus 10 reagent cassette. Reformatted interpretation section to include interferences.	U Uddayasankar	18-Dec-2018
6	Included Section 8 regarding “Unacceptable samples”. Minor changes in the 2.0 PRINCIPLE: Glucose. In section 9: Obtaining sample for in patients samples, provided a limit of 4 hours from time of collection to results. In section 10: Interpretation of Results for Glucose False positives results, contamination with oxidizing agents and detergents were added. Added the new urinalysis process implemented in Nov 2020 to reflect the removal of reflex from dipstick to microscopy. Removed Liquichek Urinalysis Controls from Section 3 Materials.	H. Vakili	22-Oct-2020
7	Update to Unacceptable Specimens (p.4). Update to Obtaining Sample on Outpatients section (p.5). Updated references (p.8).	H Vakili	8 Feb 2021
8	Addition of recommended time for refrigerated samples to return to room temperature before dipstick testing. Few formatting changes throughout the document.	H Vakili	4 Mar 2021

1.0 CLINICAL SIGNIFICANCE:

Urinalysis is routine for many patients seen in the physician's office or medical clinic, and for most physical examinations. It is one of the most useful indicators of health or disease, especially in the areas of metabolic and renal disorders.

Test results may provide information regarding the status of carbohydrate metabolism, kidney and liver function, acid-base balance, and urinary tract infections based on multiple physicochemical properties of urine to assess glucosuria, proteinuria, hematuria, leukocyturia and infection. The urine dipstick provides a rapid semiquantitative assessment of urine on a series of test pads embedded on a reagent strip to test for: Glucose, Bilirubin, Ketone, Specific Gravity, blood, pH, Protein, Urobilinogen, Nitrite and Leukocytes.

UR test code is dipstick only, with no reflex to microscopic testing. Thus, only dipstick results will be reported, regardless of positive or negative results for any components if UR is ordered.

However, when the specimen is registered with UR+URR code for Urine Renal Workup, indicating both dipstick and microscopy are requested. Then, microscopy will be performed when the dipstick is positive for blood, protein, or leukocytes.

2.0 PRINCIPLE:

Glucose: this test is based on a double sequential enzyme reaction. First enzyme, glucose oxidase, catalyzes the formation of gluconic acid and hydrogen peroxide from the oxidation of glucose. In the CLINITEK Novus 10 urinalysis cassette, the indicator reaction uses a second enzyme, peroxidase, which catalyzes the oxidative coupling of 4-amino-antipyrine and 4-methylcatechol by hydrogen peroxide The reaction of hydrogen peroxide with a potassium iodide chromogen leads to oxidized chromogen with colors ranging from green to brown.

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.

Ketone: this test is based on the development of colors ranging from buff-pink, for a negative reading, to purple when acetoacetic acid reacts with nitroprusside.

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.
In the CLINITEK Novus, specific gravity is measured using refractometry. The concentration of dissolved particles present in the solution determines the velocity and angle at which light passes through a solution. The clinical refractometer makes use of these principles of light by using a prism to direct a specific (monochromatic) wavelength of daylight against a manufacturer-calibrated specific gravity sale. The concentration of the specimen determines the angle at which the light beam enters the prism. Therefore, the specific gravity scale is calibrated in terms of the angles at which light passes throughout the specimen.

Blood: this test is based on the peroxidase-like activity of hemoglobin which catalyzes the reaction of cumene hydroperoxide and 3,3',5,5' – tetramethylbenzidine. The resulting color ranges from orange through spotty green, green to dark blue.

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 and blue.

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.

Urobilinogen: this test is based on a modified Ehrlich reaction, in which p-diethylaminobenzaldehyde in conjunction with a color enhancer reacts with urobilinogen in a strongly acid medium to produce a range from light orange to dark pink.

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)quilin-3-ol to produce a pink color.

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.

3.0 MATERIALS:

Reagents / consumables provided	**Equipment required**
• Multistix 10 SG reagent strips (Siemens HealthCare) • CLINITEK Novus 10 urinalysis cassette • BioRad qUAntify Plus Controls • Conical Centrifuge tubes	Clinitek Status, Advantus and Novus Analyzer & operator manual (Siemens HealthCare)

Urine dipstick is performed based on the instruction provided in instrument specific SOPs for urinalysis (Clinitek Novus: 110-50-17; Clinitek Advantus: 110-50-18; Clinitek Status: 110-50-19).

4.0 PRECAUTIONS FOR HANDLING MULTISTIX 10SG TEST STRIPS:

- Protect reagent strips from light, moisture, and excessive heat to prevent loss of sensitivity.
- Store at temperatures between 15-30 °C.
- Do not remove desiccant from bottle.
- Keep bottle tightly capped.
- Remove only enough reagent strips for immediate use.
- A new bottle may be used 2 months after opening. Never use the strips past the unopened expiry date.
- Never transfer strips to another bottle
- Avoid contamination of reagent strips.
- Avoid handling test end of strip.
- Do not lay reagent strip on table surface – use clean sheet of paper (not absorbent towel).
- Do not use in presence of volatile acid or alkaline fumes.

5.0 PRECAUTIONS FOR HANDLING CLINITEK Novus 10 Urinalysis Cassettes:

- Until ready to use, store the CLINITEK Novus 10 Urinalysis Cassette unopened at 15 – 30 ^{o}C. Never use cassettes past the expiry date
- After removing the cassette from the tray, it should be immediately loaded into the system
- Cassettes exposed to the outside environment for greater than 15 minutes may not yield satisfactory results.
- Cassettes must be used within 14 days after opening and loading into the system.
- Protect against expire to light, heat, and ambient moisture.

6.0 SPECIAL SAFETY PRECAUTIONS:

Use appropriate (universal) precautions in the collection, handling, storage and disposal of the specimens, used test strips and reagents. Discard used pipettes, tubes and reagent test strips in a proper biohazard container. Lab coat, gloves and eye protection should be used when handling samples.

7.0 SAMPLE REQUIREMENTS:

- Collect freshly-voided urine (10-15 mL) in a clean, dry clear container with a tight fitting lid. A first-morning specimen is preferred but random collections are acceptable. Mix the specimen just before testing and test it within 2 hours of collection for best results.
- Specimen tube must be labelled with two patient identifiers, according to Shared Health Specimen Acceptance Policy, Doc **#** 10-50-03. Do not centrifuge. **Collection date and time must be provided on the requisition, as lack of this information may lead to specimen rejection.**
- If testing cannot be done within two hours of voiding, refrigerate the specimen immediately. Refrigerated samples will be tested by dipstick only. Let the refrigerated samples return to room temperature for 30 minutes before dipstick testing.
- NO microscopy is performed on refrigerated specimens.
- Based on current CLSI guidelines and CAP requirements for optimizing the accuracy of results that both the chemistry tests are performed, and sediment examined microscopically within 2 hours of voiding.

8.0 UNACCEPTABLE SAMPLES:

- Grossly visibly bloody urines should be rejected.
- A microscopic examination will not be performed on any specimens older than 4 hours from time of void, unless collected in BD preservative tubes (Doc # 110-20-54).
- No dipstick is performed on urine specimens older than 4 hours from the time of collection unless refrigerated (up to 24 hours).
- No dipstick and microscopy is performed on urine specimens collected in BD preservative tube older than 72 hours from the time of collection (Doc # 110-20-54).
- No dipstick and microscopy is done on urine collected in UriSwab or any other preservative tubes which contain boric acid as a preservative agent. Boric acid affects a number of test strip reactions including protein, glucose and pH.
- If there is no collection time, the specimen is rejected. (HSC ONLY: exceptions would be Adult and Children's emergency after confirming with the health care team the time of collection and if the sample is not older than 4 hours from time of collection)

NOTE: Prolonged exposure at room temperature may result in microbial contamination with changes in the pH. A shift to alkaline may cause false positive protein tests. Urine with glucose may decrease in pH as organisms utilize the sugar. Bacterial growth may cause false positive blood reactions from the peroxidase produced.

9.0 OBTAINING SAMPLE:

Inpatients:

- Urine samples are collected and labeled with patient's identifiers including recorded time of void by nursing staff, and are brought to the lab for processing ideally in 2 hours and no later than 4 hours from the time of voiding.

Outpatients:

- Technologist provides the patient with a pre labeled (Name, PHIN, DOB) sample cup. After the patient is given the cup, the technologist provides collection instructions and directs patient to the washroom and where to place the collected specimen.

Note: If a Culture & Sensitivity (C&S) is ordered on a mid-stream urine (MSU) with a urinalysis, a sterile specimen container must be provided.

Patients that are unable to provide a urine sample are given a sterile specimen container and requisition to take home. **If Renal workup (UR+URR) is ordered, it is encouraged to collect the specimen at the site to ensure analysis including both dipstick and microscopy are performed within acceptable time frame of less than 4 hours from time of collection.**

Container must be labelled with patient's name, PHIN and test required (Dipstick (UR) and/or MSU / C&S). These patients are instructed to bring sample back to lab at their convenience. Patients must be instructed to keep specimen in fridge until time of arrival and specimen must be brought to lab on same day of collection with date and time of collection on label**. If samples are kept refrigerated, no microscopy will be performed.**

10.0 INTERPRETATION OF RESULTS:

Extremely dark colored or highly turbid sample may give error codes and not produce a dipstick result.

If dipstick cannot be provided due to interfering substances, delete the UR and order test code UR2 using VFR format in LIS and perform urine microscopy (SOP # 110-50-23). Comment code &URRNA must also be used to include this comment in the report "Due to interfering substances, urinalysis dipstick is unavailable. Microscopic screen added."

Glucose:
The kidney normally excretes small amounts of glucose (<1.67 mmol/L). These amounts are usually below the sensitivity of this test but on occasion may produce color between the negative and the 6 mmol/L color blocks and that is interpreted by the instrument as positive. Results at the first positive level may be significantly abnormal if found consistently. Results at the first positive should be correlated with other laboratory findings, patient history, and clinical symptoms.

Reference value: Negative

Interferences:

False Positive	*False Negative*
Urine samples with a pH of 9.0 and greater, hypochlorite, contamination with oxidizing agents and detergents	Delayed processing and prolonged exposure at room temperature; high specific gravity; ascorbic acid (>3 mmol/L); acetylcysteine; captopril; mesna; curcuma; ketone bodies

Bilirubin:
Normally no bilirubin is detectable in urine by even the most sensitive methods. Even trace amounts of bilirubin are sufficiently abnormal to require further investigation. Atypical colors may indicate bile pigment abnormalities.

Reference value: Negative

Interferences:
Atypical colors may indicate the presence of bile pigment abnormalities and the urine should be tested further.

False Positive	*False Negative*
Metabolites of Etodolac (may also cause atypical results); Indican (indoxyl sulfate); p-aminosalicylic acid; metabolites of drugs which give a color at low pH (e.g. Pyridium)	Indican (indoxyl sulfate); acetylcysteine; ascorbic acid; boric acid; hypochlorite; captopril; mesna; nitrite; curcuma; citric acid; chlorhexidine, or oxalic acid; improper storage or light exposure

Ketones:
Normal urine specimens ordinarily yield negative results with this reagent (<0.2 mmol/L). Detectable levels of ketones may occur in urine during physiological stress conditions such as fasting, pregnancy and frequent exercise. In ketoacidosis, starvation or with other abnormalities of carbohydrate or lipid metabolism, ketones may appear in urine in large amounts (>1.0 mmol/L) before serum ketone is elevated.

Reference value: Negative

Interferences:

False Positive	*False Negative*
Highly pigmented urine; levodopa metabolites; free sulfahydryl drugs (mesna, captopril, N-acetyl cysteine); curcuma; formalin; imipenem; hydrochlorothiazide	Boric acid; formalin; hypochlorite; meropenem; Lodine

Specific Gravity:
Random urines may vary in specific gravity with an approximate normal range of 1.005 – 1.030. If the SG of random urine is ≥ 1.023, the concentrating ability of the kidneys can be considered normal. Results should be correlated with other laboratory findings, patient history, and clinical symptoms.

Reference Interval: 1.005 – 1.030

Interferences:
The Multistix test is not affected by radiopaque dyes, while those measured with a refractometer are affected.

False Positive	*False Negative*
Multistix: Moderate quantities of protein Refractometer: Dextran solutions, IV radiopaque dyes, proteinuria.	Multistix: Highly buffered alkaline urines.

Blood:
Normally, no hemoglobin is detectable in urine (<3 RBC/uL). The significance of the “trace reaction” may vary among patients, and clinical judgment is required for assessment in an individual case. Green spots (intact erythrocytes) or green color (free hemoglobin/myoglobin) on the reagent area within 50 seconds indicates a need for further investigation. Blood is often, but not always, found in urine of menstruating females.

Reference value: Negative

Interferences:
Lysed erythrocytes may cause discrepancies with microscopy.

False Positive	*False Negative*
Peroxidases (e.g. microbial); strong oxidizing agents (e.g. hypochlorite)	Captopril and other sulfhydryl compounds; acetylcysteine; ascorbic acid; formalin; quinidine; cefoxitin; levodopa; mesna; Keflin; curcuma; Lodine; hydrochlorothiazide; metformin; chlorhexidine; chloroquine.

pH:
Reference range: 5.0 to 8.0

Interferences:
Bacterial growth may cause a marked alkaline shift (pH>8.0) because of urea conversion to ammonia if specimen is left at room temperature for more than 4 hours.

Protein:
Normally, no protein is detectable in urine, although the normal kidney excretes a minute amount (<0.15 g/L). Clinical proteinuria is indicated at greater than 0.5 g protein per day (>0.3 g/L). Clinical judgement is needed to evaluate the significance of Trace results. 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.

Reference value: Negative

Interferences:

False Positive	*False Negative*
Highly buffered or alkaline urine (pH>9.0); prolonged exposure at room temperature due to change in the pH; quinidine; chlorhexidine; chloroquine; Iodine; hemoglobin; pigmented specimens; quaternary ammonium compounds; high specific gravity	Primary protein is not albumin (e.g. Bence-Jones protein, curcuma.)

Urobilinogen:
Normally present in urine up to 16 umol/L. A result of ≥33 umol/L should be correlated with other laboratory findings, patient history, and clinical symptoms. Evaluation of both the bilirubin and urobilinogen results helps in the differential diagnosis of jaundice, as well as other liver and biliary disorders.

Reference range: 3 – 16 µmol/L

Note: *Not a reliable test for porphobilinogen.*

Interferences:

False Positive	*False Negative*
Elevated nitrite levels; phenazopyridine; any other Ehrlich's reactive substance (porphobilinogen, indicans); atypical colours caused by sulfonamides; p-aminobenzoic acid; p-aminosalicylic acid; beet ingestion; methyldopa; procaine; chlorpromazine	Formalin; improper storage; acetylcysteine; captopril; hypochlorite; mesna; Tagamet; curcuma; Lodine; sulfamethoxazole; chlorhexidine; glucose; hydrochlorothiazide; lactose; meropenem; or nitrofurantoin.

Nitrite:
Normally no nitrite is detectable in urine. Many enteric gram-negative organisms give positive results when their number is greater than 10^5/mL (>16.2 umol/L nitrite). A minimum of 4 hours of bladder incubation significantly increases the likelihood of obtaining a positive result.

Reference value: Negative

Interferences:

False Positive	*False Negative*
Highly colored substances; curcuma or beet ingestion; improper storage with bacterial proliferation	High specific gravity; ascorbic acid; oxalic acid; Lodine; formalin; chlorhexidine; various factors that inhibit or prevent nitrite formation despite bacteruria (e.g. nitrate reductase negative bacteria, lack of urine nitrate, presence of antibiotics, insufficient time for bacteria to reduce nitrate, or large quantities that convert nitrite to nitrogen).

Leukocytes:
Normal urine samples generally yield negative results. An increase in leukocytes (≥ 10 leukocytes/uL) may be an indication of urinary tract infection. Increased leukocytes may also be present in noninfective conditions. A result greater than Trace may require follow-up testing and should be correlated with other laboratory findings, patient history, and clinical symptoms.

Reference value: Negative

Interferences:
Detects esterase activity from either intact or lysed granulocytic leukocytes. Lysed granulocytic leukocytes may produce apparent discrepancies between positive dipstick and negative microscopic results. Lymphocytes do not produce a positive reaction.

False Positive	*False Negative*
Highly colored substances; vaginal contamination of urine; formalin; curcuma	High specific gravity; glycosuria; ketonuria; proteinuria; oxalic acid; ascorbic acid; boric acid; strong oxidizing agents; quinidine; Tagamet; glycine; chloroquine; sulfamethoxazole; chlorhexidine; nitrofurantoin; Lodine; drugs such as tetracycline, gentamicin, and cephalosporin.

11.0 REFERENCES:

- Siemens Clinitek Status Urinalysis Operating Manual
- Siemens Multistix 10 SG Reagent Strips
- Siemens CLINITEK Novus 10 Urinalysis Cassette
- Shared Health document **#** 10-50-03**,** Specimen Acceptance Policy
- Shared Health document # 110-10-09, Quality Control of Urinalysis Test Strips
- GP16-A2 Vol. 21 No. 19 Urinalysis and Collection, Transportation, and Preservation of Urine Specimens; Approved Guideline—Second Edition
- **CLSI document, GP16-A3:** Urinalysis and collection, transportation, and preservation of urine specimens. Approved Guideline – 3rd Edition Feb 2009
- Joris Delanghe and Marijn Speeckaert, Preanalytical requirement of urinalysis, Biochemia Medica 2014;24(1): 89-104.