



# Multistix<sup>®</sup> 10 SG • Multistix<sup>®</sup> 9 • Multistix<sup>®</sup> 9 SG • Multistix<sup>®</sup> 8 SG • Multistix<sup>®</sup> 7 • N-Multistix<sup>®</sup> SG • N-Multistix<sup>®</sup> • Multistix<sup>®</sup> SG • Multistix<sup>®</sup> • Bili-Labstix<sup>®</sup> 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:<sup>1-3</sup>

- 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.<sup>1,4</sup>

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<sup>®</sup> 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.<sup>5</sup>

**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.**

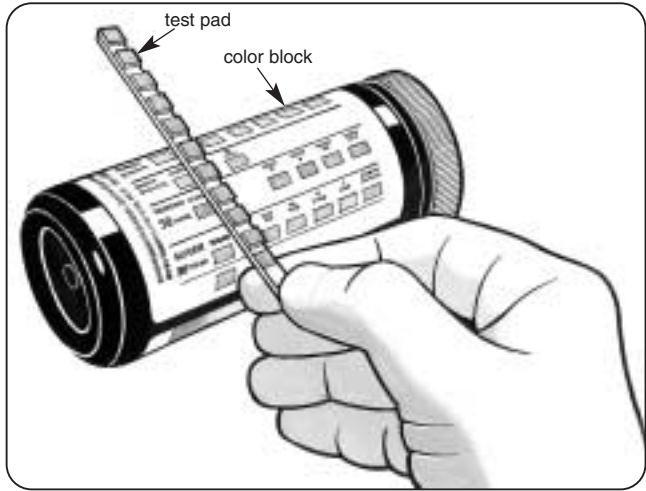


3. **Drag** the *edge* of the strip against the container rim to remove excess urine.



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.



4. 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<sup>®</sup> 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 (Macrochantin, 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  $\geq 30$  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.<sup>1,6-7</sup> 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.<sup>8</sup>

**Limitations:** A visibly bloody urine may cause falsely elevated results.<sup>8</sup>

#### BLOOD:

**Expected values:** Normally, no hemoglobin is detectable in urine (<0.010 mg/dL or 3 RBC/ $\mu$ L). 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.<sup>1,9</sup>

**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.

**Limitations:** Capoten (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 ( $\geq 10$  leukocytes/ $\mu$ L) 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.<sup>1</sup> 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.<sup>1</sup> 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 ( $\geq 3$  g/dL) may cause decreased test results. The presence of cephalixin (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).<sup>2,9</sup>

**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.<sup>2,3</sup>

**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.<sup>1</sup>

**Sensitivity:** 5–10 mg/dL acetoacetic acid

**Performance characteristics:** The test reacts with acetoacetic acid in urine. It does not react with acetone or  $\beta$ -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.<sup>1</sup>

**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.<sup>1</sup>

**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  $\geq 6.5$ . Strips read instrumentally are automatically adjusted for pH by the instrument. The Bayer SG test is not affected by the presence of radioactive 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.<sup>1</sup> 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<sup>®</sup> 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 Iodine (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.<sup>1</sup>

**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°–26°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 tetrabromophenol 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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