

AdultaCheck® 4

Adulteration Detection on a Dipstick with tests for Creatinine, Nitrite, Glutaraldehyde and pH

U. S. and Foreign Patents Pending
Product Code 300

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INTENDED USE

For the visual determination of creatinine, nitrite, glutaraldehyde and pH values in urine to assess the integrity of urine samples prior to Drugs-Of-Abuse Urine (DAU) testing, in order to determine if the sample has been adulterated. The AdultaCheck® 4 is not for use in diagnosing disease or illness to effect treatment, and/or cure; it is For Forensic / Toxicology Use Only.

SUMMARY

AdultaCheck® 4 (25 test strips per bottle) are reagent dipsticks with reagent impregnated pads for 4 distinct and specific analytes. The pads provide tests for creatinine, nitrite, glutaraldehyde and pH on urine submitted for DAU Testing.

Please refer to the carton and bottle label for specific reagent areas on the AdultaCheck® product you are using.

The tests provide information on the possible adulteration of urine samples submitted for DAU testing. In drugs-of-abuse screening, it is important to insure the integrity of samples. Simple dilution of urine is probably the most common form of urine adulteration. Urine creatinine values and appearance are useful as an adjunct in detecting *in vivo* and *in vitro* dilution of urine samples.^{1,2,3}

Tests for pH and creatinine can aid in the detection of common methods for defeating urine drug tests including dilution, or adulteration of the sample with bleach, vinegar, Visine, sodium bicarbonate, Drano, soft drinks or hydrogen peroxide. Since tests for pH and creatinine detect many of these common adulterants, some drug users are utilizing commercially available adulterants, including UrinAid and INSTANT CLEAN ADD-IT-IVE. The active ingredient in these products is an aldehyde (glutaraldehyde). The Roche RIA Abuscreen™ assay is not significantly affected by glutaraldehyde, however, this ingredient does inactivate the enzymes used in all Syva EMIT™ assays, BMC CEDIA™ assays, and the Roche ONLINE™ Cocaine assay, causing false negatives. The Roche ONLINE™ THC assay is also subject to false positive results due to the presence of glutaraldehyde. These results have been documented by independent research.^{4,5}

Positive nitrite test results obtained on urine specimens usually depend on the conversion of nitrate (derived from the diet) to nitrite in the urine by the action of some Gram-negative bacteria.^{9,10} In the

absence of infection, normal urine is negative. Now, published reports indicate that nitrite is being used as an adulterant in urine samples submitted for Drugs-of-Abuse testing.^{6,13} The Nitrite assay will detect addition of nitrite above clinical levels.

The AdultaCheck® 4 reagent strips are ready to use, single-use test strips and have proven to be an accurate, cost-effective, and efficient test for the detection of adulterated samples at the point of collection.^{11,14} Dispose of each strip after the evaluation of test results. The strips are read visually by comparison of results on the test pads with the color chart provided on the bottle label. Directions must be followed precisely using accurate timing for optimum results. The reagent strips must be kept in the bottle, tightly closed, to maintain reagent activity. For optimum results use fresh, well-mixed uncentrifuged urine samples.

PRINCIPLE

Creatinine: In this reaction, creatinine interacts with a creatinine indicator in an alkaline medium and forms a blue / purple-colored complex. Urine creatinine values are useful as an adjunct in detecting adulteration of urine samples submitted for drugs-of-abuse testing.^{1,2,3,14} The color intensity is directly proportional to the concentration of creatinine and is observed visually and compared to the color chart on the bottle to obtain a result.

Nitrite: This assay detects nitrite. Nitrite (if present) reacts with the reagent's aromatic amine on the test pad to form a diazonium salt, which couples with an indicator to yield a color complex.^{9,10} The color is observed visually and compared to the color chart on the bottle to obtain a result. Urines adulterated with nitrite will yield a dark purple color. The measurement of urine nitrite values in excess of 150 mg/L are useful as an adjunct in detecting adulteration of urine samples submitted for drugs-of-abuse testing.^{6,13,14,15}

Glutaraldehyde: When urine containing glutaraldehyde contacts the reagent pad on the dipstick, an aldehyde indicator present reacts with the aldehyde group of the glutaraldehyde to form a colored product. The reaction produces a color that is observed visually and compared to the color chart on the bottle to obtain a result. The detection of any glutaraldehyde in urine is indicative of adulteration.^{4,5,14}

pH: The analysis is based on an indicator principle that gives a change in color throughout the urinary pH range. A sample of urine is added to the test pad and, depending on the hydrogen ion concentration of the urine, the indicator will exhibit a change in color. This color change is observed visually and compared to the color chart on the bottle to determine a pH value. Published studies have shown that sample pH can have a significant effect on the results of drug screening tests.^{14,16,17,18}

REAGENTS

Creatinine: 0.91% creatinine reactive indicator, 0.08% alkaline buffer, and 99.1% non-reactive ingredients.

Nitrite: 12.4% w/w nitrite reactive indicators, 10.8% w/w buffer and 76.8% w/w non-reactive ingredients.

Glutaraldehyde: 0.006% aldehyde indicator, 0.8% buffer, and 99.19% non-reactive ingredients.

pH: 0.6% w/w indicators and NaOH/HCl to adjust pH, and 99.4% non-reactive ingredients.

WARNINGS AND PRECAUTIONS

AdultaCheck® 4 reagent strips are for Forensic / Toxicology use only. Handle all urine specimens as if potentially infectious. Handle the reagent strips with care. Avoid contact with skin and mucous membranes.

STORAGE

Store AdultaCheck® 4 reagent strips at room temperature between 15°C-30°C (59°-86°F). Do not use product past the expiration date.

RECOMMENDED PROCEDURES FOR HANDLING ADULTACHECK® 4 REAGENT STRIPS

All unused strips must remain in the original bottle. Do not remove desiccant(s) from bottle. Open the bottle and remove strip just prior to use and close bottle tightly after removing the strip. Do not allow the test areas of the reagent strip to come in contact with any surface or liquid except the urine sample to be tested.

For legal, forensic purposes it is recommended that AdultaCheck® 4 strips not be dipped into the primary collection container. To avoid any question of sample contamination, a small portion of the specimen should be poured into a second container for use with the AdultaCheck® 4, or poured directly onto the test strip.

SPECIMEN COLLECTION AND PREPARATION

- * Urine samples should be collected in clean glass or plastic containers.
- * Do not centrifuge or add preservatives to the urine specimen.
- * Test urine specimen as soon as possible after collection. If the urine specimen cannot be tested within one hour, the specimen should be refrigerated at 2-8 °C immediately.
 - Bring refrigerated specimen to room temperature and mix
 - thoroughly before testing.
- * Handle and dispose of all human urine samples as if potentially infectious.

PROCEDURE

1. Collect fresh urine specimen in a clean, dry container. Mix well immediately prior to testing.
2. Remove one strip from the bottle and replace the cap.
3. Completely immerse reagent areas of the strip in the urine and remove it immediately to avoid dissolving out reagents. As it is being withdrawn from the urine specimen, run the edge of the strip against the container to remove excess urine.
4. Hold the strip in a horizontal position, test pads up, to prevent possible mixing of chemicals from adjacent reagent areas and / or contaminating hands with urine.
5. Visually compare the reagent pads on the strip to the corresponding blocks on the Color Chart located on the bottle label. Read results at the times indicated.
6. Hold the reagent strip close to the Color Chart and carefully match

the colors on the strip with those on the Color Chart blocks and record the results of each test. Avoid touching the strip to the label as this may contaminate the Color Chart with color from the reagent strip. Proper read times are critical for optimal results. The read time listed on the bottle label for each test is to begin as soon as the strip has been withdrawn from the specimen and held in a horizontal position.

QUALITY CONTROL

For best results, performance of the reagent strips should be confirmed whenever a new bottle is opened by testing with known normal and abnormal controls. Chimera recommends its AdultaCheck® Quality Control Set for this purpose. Each laboratory should establish its own goals for adequate standards of performance and should question handling or testing procedures if these standards are not met.

RESULTS

Results with AdultaCheck® 4 Reagent Strips are obtained directly from the Color Chart by visual comparison.

LIMITATIONS

The object of tests for adulteration is to discover deviations in urine samples, such as dilution, or the addition of drug-test interfering substances. The list of limitations below includes those compounds or physical properties that may affect the test. Medications may cause abnormal results due to discoloration of the urine, and consequently mask the reagent pad color reaction.

Creatinine: The list of substances and conditions that may affect creatinine levels *in vivo* are given by Young et al.⁷ and Friedman et al.⁸ No representation is made by manufacturer regarding the completeness of these lists or accuracy of the information contained therein.

Nitrite: The test is specific for nitrite. False-negatives may result from extremely high ascorbic acid levels (> 1000 mg/dL) or contact with hypochlorite (in containers).

Glutaraldehyde: This assay is for the presence of aldehydes. Normal endogenous urine constituents (glucose, protein, etc.) do not interfere with this assay. Extremely high levels of urinary ketone bodies (above 75 mg/dL) may effect a false positive result.

pH: The test allows measurement of urine pH ranging from pH 3.0 to pH 9.0. There are no known interference's with this test.

EXPECTED VALUES

Creatinine: In a study by manufacturer, on 40 random urine samples, values were obtained which ranged from 22.8 to 246.7 mg/dL, with a mean value of 122.4 mg/dL. Needleman¹¹, et. al., in a study of 350 single collection random samples, reported values ranging from 25 to 300 mg/dL. Results above 300 mg/dL were listed only as 300 mg/dL. Twenty-six specimens had values of at least 300 mg/dL. Of the 350 specimens, 340 had values greater than 30 mg/dL. The mean creatinine concentration for the 350 samples was 171.6 mg/dL. Goldberger³, et al, in analyzing 1206 individual specimens, reported

values ranging from 18.1 to 531.7 mg/dL with a mean value of 159.5 mg/dL. Three of the specimens had creatinine values of less than 20 mg/dL.

The DOT states that urine specimens with creatinine values of less than 20 mg/dL, and specific gravity of less than 1.003, suggest adulteration.

Values of less than 20 mg/dL may reasonably be suggestive of adulteration¹². As the creatinine value approaches 0 mg/dL, the more suspect the specimen becomes.

The ranges should serve only as a guideline,^{1,2,3,11,12} and since they are affected by patient age, sex, diet, and local population variations, it is recommended that each laboratory establish its own range of expected values.

Nitrite: Nitrite is not normally found in random urine. The presence of 0.1 mg/dL nitrite may indicate the presence of gram neg. bacteria.^{9,10} Nitrite levels due to urinary tract infections or bacterial contamination and improper storage, might not exceed 36 mg/L.¹³ In this assay 1.0 to 50.0 mg/L is considered "normal". The positive cutoff concentration for this assay is 150.0 to 500.0 mg/L. Any result at or above this value may indicate adulteration. Adulterated samples may yield results of >1900 mg/L nitrite.^{6,13,14,15}

Glutaraldehyde: Glutaraldehyde is not a normal constituent of human urine.^{4,5,14} Acute ketosis, in a patient, may cause a false positive result. The strip, therefore, indicates the presence or absence of the adulterant, glutaraldehyde. The positive cutoff for this assay is 0.20% to 0.40% vol/vol.

pH: Normal random urine pH ranges from pH 4.5 to 8.0.⁹ Values below pH 4.5 or above pH 8.0 may be suggestive of adulteration.^{14,16,17,18}

SAMSHA* recommended cutoffs for the specific analytes are:

creatinine: < 20 mg/dL

nitrite: ≥ 500 mg/L

glutaraldehyde: the presence of any exogenous substance is considered adulterated

pH: ≤ 3 or ≥ 11

* Substance Abuse and Mental Health Services Administration

PERFORMANCE CHARACTERISTICS

The user should be aware that color perception depends upon several external factors like the quality and amount of available light and the visual capability of the user (a color-blind person may need assistance).

Creatinine: The creatinine test permits determination of urinary creatinine between 0 and 400 mg/L. While urinary creatinine may vary at the upper end, the lower level is usually around 20 mg/dL creatinine. Samples with less than 20 mg/dL creatinine should be considered abnormal, and possibly adulterated.^{1,2,3,12,14} A sample with less than 10 mg/dL creatinine may be considered substituted.

Nitrite: The nitrite test permits determination of nitrite concentration in urine from negative to 10000.0 mg/L in increments from negative

to 150.0, 500.0, 1000.0, and 10000.0 mg/L. Nitrite is not normally found in random urine. Values of 1.0 - 50.0 mg/L may indicate a urinary tract infection or bacterial growth following improper storage. In this assay, if nitrite is present above 150.0 mg/L, it is an indication of adulteration of the urine sample.

Glutaraldehyde: The presence of any glutaraldehyde in a urine specimen is indicative of adulteration. The test is for aldehydes that are not found in normal urine.

pH: The pH test permits determination of pH concentration in urine from a pH of 2.0 to 12.0 in increments of 1 pH unit. Urinary pH varies in normal specimens from pH 4.5 to 8.0.⁹ Samples with pH outside this range should be considered adulterated.^{14,16,17,18}

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