One Step COC Test Kit

For Urine

(Revised Jan. 30th, 2001)

Intended Use

The one step COC (Cocaine) test is a simple one step immunochromatographic assay for the rapid, qualitative detection of COC and benzoylecgonine as the primary metabolite in urine. The cutoff of the test is 300 ng/ml of COC. It is the same as the SAMHSA recommended assay cutoff.

The COC test provides only a preliminary analytical result. A more specific alternative chemical method must be used in order to obtain a confirmed analytical result. Gas chromatography, mass spectrometry (GC/MS) is the preferred method. Clinical consideration and professional judgment should be applied to any drug of abuse test result, particularly when preliminary positive results are used.

Explanation of the Test

Cocaine, derived from the leaves of the coca plant, is a potent central nervous system stimulant and a local anesthetic. Cocaine induces euphoria, confidence and a sense of increased energy in the user; these psychological effects are accompanied by increased heart rate, dilation of the pupils, fever, tremor and sweating. Cocaine is used via smoking, intravenous, intranasal or oral administration, and is excreted in the urine primarily as benzoylecgonine in a short time. Benzoylecgonine has a longer biological half-life (5-8 hours) than cocaine (0.5-1.5 hours) and can generally be detected for 24-80 hours after cocaine use or exposure.

The COC test is based on the principle of highly specific immunochemical reactions between antigens and antibodies, which are used for the analysis of specific substances in urine. Major antibodies and buffers used in the COC Test Kit are listed as the following; Goat anti-rabbit IgG (Control Line), mouse monoclonal antibody against COC (Gold Conjugate), BSA-COC (Test Line) and phosphate buffer and tris buffer. The cutoff of the test is 300 ng/ml of COC.

The COC test cassette has a letter T and C as the "Test Line" and the "Control Line" on the surface of the case. Both the "Test Line" and "Control Line" in the result window are not visible before applying any samples. The "Control Line" is used for procedural control. The control line should always appear if the test procedure is performed properly and the test reagents are working properly.

Materials Provided

The COC test kit contains the following items to perform the assay:

1. COC test device.
2. Disposable sample dropper.
3. Instructions for use.

Materials Required But Not Provided

1. Specimen collection container.
2. Clock or timer.

Precautions

1. For professional in vitro diagnostic use only.
2. Avoid cross contamination of urine samples by using a new urine specimen container and dropper for each urine sample.
3. Urine specimens are potentially infectious. Proper handling and disposal methods should be established according to good laboratory practices.
4. Do not eat or smoke while handling specimens in the laboratory.
5. COC device should remain in its original sealed pouch until ready for use.
6. Do not use the test if the pouch is damaged or the seal is broken.
7. Do not use the test kit after the expiration date.

Storage and Stability

COC test kit should be stored at 4-30 °C in the original sealed pouch. The expiration date given was determined under normal laboratory conditions.

Specimen Collection and Preparation

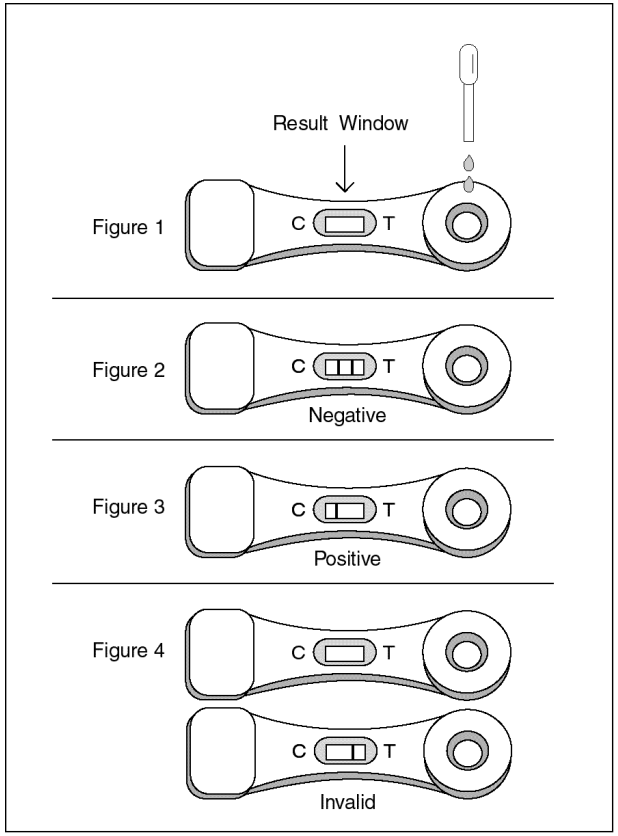
1. Fresh urine specimens do not require any special handling or pretreatment.
2. Specimens should be collected in a clean glass or plastic container.
3. If testing will not be performed immediately, specimens should be refrigerated.
4. Specimens should be brought to room temperature before testing.
5. Specimens containing precipitate may yield inconsistent test results. Such specimen must be clarified prior to assaying.

Procedure of the Test

1. Remove the test disk from the foil pouch, and place it on a flat, dry surface.
2. Holding the sample dropper above the test disk. Squeeze 2 drops of specimen into the sample well (See following Figures)
3. Interpret the test results at 3 to 5 minutes.

Interpretation of Results

1. As the test kit begins to work, a purple band will appear at the left section of the result window to show that the Control Line is working properly.
2. The right section of the result window indicates the test results. If another purple band appears at the right section of the result window, this band is the Test Band. (Figure 1)



Negative: Two Color Bands

The appearance of two purple bands within the result window indicates a negative test result. No COC above the cut-off level has been detected. The color of the Test band may be lighter or darker than that of the Control Band. (Figure 2)

Positive: One Color Band

The appearance of only one purple band within the result window indicates the result is positive, i.e. the specimen contains COC at a concentration above the cut-off level.
The urine specimen should be confirmed with a more specific alternative method such as GC/MS, before a positive determination is made. (Figure 3)

Invalid:

A distinct purple band should always appear in the left section of the result window. The test is invalid if no color band forms in the left section of the result window. (Figure 4)

Note. A very faint band in the right section of the result window, visible in 5 minutes, indicates that the amount of COC in the sample is near or below the cut-off level of the test. The urine specimen should be retested, or confirmed with a more specific alternative method such as gas chromatography/mass spectrometry, before a positive determination is made.

User Quality Control

Control standards are not supplied with this kit; however, it is recommended that a control be tested as good laboratory testing practice. For information on how to obtain controls, contact Technical Service. Before using a new kit with patient specimens, positive (cutoff and 25% more than cutoff level) and negative (25% below cutoff level) controls should be tested to confirm the test procedure, and to verify the tests produce the expected Q.C. results.

Limitations

1. The test is designed for use with unadulterated human urine only.
2. There is a possibility that factors such as technical or procedural errors, as well as other substances in the urine samples may interfere with the test and cause erroneous results.
3. Adulterants, such as bleach and/or alum, in urine specimens may produce erroneous results regardless of the method of analysis. If adulteration is suspected, the test should be repeated with a new sample.
4. A positive test result does not provide any indication of the level of intoxication or urinary concentration.
5. The test results read after 5 minutes may not be consistent with the original reading obtained within the 5 minutes reading period. The test must be read within 5 minutes of sample application.
6. Food and tea containing poppy products and/or coca leaves may produce a positive result.

Expected Values

The COC test is a qualitative assay. The amount of drugs and metabolites present in the urine cannot be estimated by the assay. The assay results distinguish positive from negative samples. A positive result indicates the sample contains COC above the cut-off concentration.

Performance Characteristics and Comparison Studies

The COC test has been shown to detect an average of 300 ng/ml or more of COC metabolites in urine. The accuracy of the COC was evaluated in comparison to a commercially available immunoassay. A total of 50 negative real urine patients' samples (concentration of COC range of 0-220 ng/ml) and 50 positive real patients urine sample (concentration of COC range of 300-1200 ng/ml) were tested by both procedures. Complete agreement was observed in 100% of the samples. All positive and part of negative urine samples were confirmed by GC/MS.

Precision and Reproducibility Studies

The precision of the COC assay was determined by carrying out the test with serially spiked COC urine samples. The four concentrations, at 0 ng/ml, at –25% from the cutoff (225 ng/ml), at the cutoff (300 ng/ml) and +25% from the cutoff (375 ng/ml) had been tested to challenge the precision of the test device. A total of 50 tests were run at 0 concentration, 50 tests at 225 ng/ml, 200 tests at 300 ng/ml and 50 tests at 375 ng/ml. A Total of 350 tests were tested. About 99% of the samples containing drug concentrations at or more than 25% over the cut-off level consistently showed positive results.

The reproducibility studies were carried out at three different sites. The urine samples contain 0, 300 ng/ml and 900 ng/ml of COC had been tested by a total of 360 COC test kits. The samples were tested two times in the same day, and in two different assays, each day for 20 days. This permits separate tests of between-days, between-assay and within day show consistent results.

Cutoff Studies

There are total of 200 urine samples including 50 samples containing zero, 50 samples below the cutoff (225 ng/ml), 50 samples at the cutoff (300 ng/ml) and 50 samples above the cutoff (375 ng/ml). Both the dBest and a commercially available immunoassay test kit tested all 200 urine samples. Complete agreement was observed as 99.43% and the test cutoff is 300 ng/ml of COC.

Specificity and Interference Studies

The following table lists compounds that are detected by the COC test. The results are expressed in terms of the concentration required to produce a positive result.

Compound	Conc. (ng/ml)
Benzoylecgonine	300
Cocaine	300
Ecgonine HCL	1000

Potentially interfering chemicals such as pain medications (Acetaminophen, 20 mg/dl), protein (2000 mg/dl), glucose (2000 mg/dl), hemoglobin (500 mg/dl) and pH of 6.0, 7.0 and 8.0 were supplemented to normal urine specimens devoid of cocaine. The test was consistently negative results. The base line urine with 300 ng/ml cocaine scored consistently positive.

References

1. Urine Testing for Drugs of Abuse, National Institute for Drug Abuse (NIDA), Research Monograph 73, 1986.
2. Ambre, J.J. Anal. Toxicol. 9: 241-5, 1985.