Buprenorphine is a potent analgesic often used in the treatment of opioid addiction. The drug is sold under the trade name Subutex®. Buprenorphine and Nortbuprenorphine in urine may be below 1 ng/mL after therapeutic administration, but can range up to 20 ng/mL in abusive situations. The plasma half-life of Buprenorphine is 2–3 hours. Complete elimination of a single-dose of the drug can take longer than 6 days, the detection window for the parent drug in urine is thought to be approximately 3 days. The Cozart® Rapid Urine BUP Test is a rapid urine screening test that can be performed without the use of an instrument. The test utilizes a monoclonal antibody to selectively detect elevated levels of Buprenorphine in urine. The Cozart® Rapid Urine BUP Test yields a positive result when the Buprenorphine in urine exceeds 10 ng/mL.

### PRINCIPLE

The Cozart® Rapid Urine BUP Test is an immunoassay based on the principle of competitive binding. Drugs which may be present in the urine specimen compete against the drug conjugate for binding sites on the antibody.

During testing, a urine specimen migrates upward by capillary action. Buprenorphine, if present in the urine specimen below 10 ng/mL, will not saturate the binding sites of antibody-conjugated particles in the test. The antibody-conjugated particles will then be captured by immobilized Buprenorphine conjugate and a visible colored line will show up in the test line region. The control line will not form in the test line region if the urine specimen contains drug concentrations above 20 ng/mL because it will saturate all the binding sites of anti-Buprenorphine antibodies. A drug-free urine specimen will not generate a colored line in the test line region because of drug specificity; while a drug-negative urine specimen or a specimen containing a drug concentration lower than the cut-off will generate a line in the control line region.

To serve as a procedural control, a colored line will always appear at the control line region, indicating that proper volume of specimen has been added and membrane/membrane has worked.

The test contains mouse monoclonal anti-Buprenorphine antibody-coated pellets and Buprenorphine-protein conjugate. A goat antibody is employed in the control line region.

### LIMITATIONS

- **For professional in-vitro diagnostic use only. Do not use after the expiration date.**
- The test should remain unopened until just before use.
- All specimens should be considered potentially hazardous and handled in the same manner as an infectious agent.
- The test should be discarded according to local regulations.

### STORAGE AND STABILITY

Store as packaged in the sealed pouch either at room temperature or refrigerated (2-30°C). The test is stable through the expiration date on the sealed pouch. The test must remain in the sealed pouch until use. DO NOT FREEZE. Do not use beyond the expiration date.

### SPECIMEN COLLECTION AND PREPARATION

The urine specimen must be collected in a clean and dry container. Urine collected at any time of the day may be used. Urine specimens exhibiting visible precipitates should be filtered, filtered, or allowed settle to obtain a clear specimen for testing.

### Urine Storage

Urine specimens may be stored at 2-8°C for up to 48 hours prior to assay. For prolonged storage, specimens may be frozen and stored below -20°C. Frozen specimens should be thawed and mixed before testing.

### DIRECTIONS FOR USE

**Allow the test, urine specimen, and or controls to reach room temperature (15-30°C) prior to testing.**

1. Place the test device on a clean and level surface. Hold the dropper vertically and transfer 3 full drops of urine (approx. 100 μL) to the specimen well (S) of the device, and then start the timer. Avoid trapping air bubbles in the specimen well. Use the illustration below to determine the volume.
2. Wait for the color(s) to appear. Read results at 5 minutes. Do not interpret the result after 10 minutes.

### INTERPRETATION OF RESULTS

**NEGATIVE:** Two distinct colored lines appear: one in the control line region (C), and another apparent colored line should be in the test line region (T). This negative result indicates that the Buprenorphine concentration exceeds the detectable level (10ng/mL).

**POSITIVE:** One colored line appears in the control region (C). No line appears in the test line region (T). This positive result indicates that the Buprenorphine concentration is below the detectable level (10ng/mL).

**LIMITATIONS**

- It is possible that the test will indicate a false negative result of the test if there is insufficient proportion of the urine specimen.
- The test method must be used to obtain a confirmed result. Liquid chromatography/mass spectrometry (LC/MS) is the analytical method used. If adulteration is suggested, the test should be repeated with another urine specimen.
- A positive result indicates presence of the drug or its metabolites but does not indicate level or intoxication, administration route or concentration in urine.
- A negative result may not necessarily indicate drug-free urine. Negative results can be obtained when drug is present but below the cut-off level.
- Test does not distinguish between drugs of abuse and certain medications.

### PERFORMANCE CHARACTERISTICS

A correlation study was conducted on fifty-eight (58) clinical specimens from patients reporting Buprenorphine use and one-hundred fifty (150) urine specimens collected from non-drug using norm. Using the Cozart® Rapid Urine BUP Test, the specimens were tested and compared to the self-reported use of Buprenorphine. All specimens, including the ones showing negative results were confirmed by LC/MS. The following results were tabulated:

### QC CHART

<table>
<thead>
<tr>
<th>Compound</th>
<th>Concentration (ng/mL)</th>
<th>Method</th>
<th>Texas</th>
<th>Total</th>
<th>Texas</th>
<th>Total</th>
<th>% Agreement</th>
</tr>
</thead>
<tbody>
<tr>
<td>Buprenorphine</td>
<td>10</td>
<td>Cozart® BUP</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100%</td>
</tr>
<tr>
<td>Buprenorphine</td>
<td>20</td>
<td>Cozart® BUP</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100%</td>
</tr>
<tr>
<td>Nortbuprenorphine</td>
<td>0</td>
<td>Cozart® BUP</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100%</td>
</tr>
</tbody>
</table>

### Complex-Reactive Compounds

The pH of an aliquoted negative urine pool was set at 5 to 9 in 1 pH unit increments and spiked with Buprenorphine 5 ng/mL and 20 ng/mL. The spiked, pH-adjusted urine was tested with the Cozart® Rapid Urine BUP Test in duplicate. The results demonstrate that varying ranges of pH do not interfere with the performance of the test.

A study was conducted to determine the cross-reactivity of the test with compounds in either drug-free urine or Buprenorphine positive urine. The following compounds show no cross-reactivity when tested with the Cozart® Rapid Urine BUP Test.

### BIBLIOGRAPHY

2. Heids IE, CN TOSCHOP. Drug Testing for Drugs of Abuse (NIDA), Research Monograph 73, 1999
3. CBI, REFC 2.0 (1995). Oxfordshire, CH41 4BY

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