ALPHA DIAGNOSTIC INTERNATIONAL

Fentanyl Rapid Test (Urine/Cassette)

A one step test for the qualitative detection of Fentanyl and its metabolites in urine. ***For Forensic Use Only.***

Summary

Fentanyl, a synthetic opiate, is a μ-receptor agonist analgesic that is 50-100 times more potent than morphine and has a short duration of action. Fentanyl, originally for chronic pain, has increased in illicit street use leading to tens of thousands of deaths in the US every year. There are several analogues and derivatives of fentanyl which are also abused and have higher potencies. Fentanyl is rapidly metabolized by the liver to the inactive metabolites norfentanyl, hydroxyfentanyl, and hydroxynorfentanyl. Fentanyl is excreted in urine with roughly 0.4-6% of the drug unchanged, 26-55% excreted as norfentanyl, and unknown amounts of hydroxyfentanyl and hydroxynorfentanyl.

Principle of the Test

The fentanyl rapid test is a qualitative, competitive test for the detection of Fentanyl and its metabolites in urine. The membrane is pre-coated with Fentanyl on the test line region of the strip and antibody coated particles. Urine migrates upward by capillary action which causes the antibody coated particles to bind to the immobilized Fentanyl resulting in a red colored line in the test region.
Norfentanyl, if present in urine at 60 ng/ml or higher will saturate the antibody coated particles resulting in no test line. A Fentanyl positive specimen will not generate a line in the test region (T) while a Fentanyl negative specimen will generate a line in the test region (T).

Precautions

- For *Forensic Use Only*. Do not use after the expiration date.
- Do not eat, drink, or smoke in the area where the specimens or kits are being handled.
- Wear protective clothing such as disposable gloves when specimens are being tested.
- Handle all specimens as if they contain infectious agents. The cassette should be disposed according to federal, state, and local regulations.
- Humidity and temperature can adversely affect results.
- Use the test within 30 minutes of opening the sealed foil pouch.

Storage and Stability

The kit can be stored at room temperature or refrigerated (2-30ºC/35-86ºF). **DO NOT FREEZE.** The test strips must remain in the sealed pouch until use. The kit is stable until the date printed on the pouch.

Materials

Materials provided: Test cassettes and transfer pipette
Materials required but not provided: Urine collection container and a timer

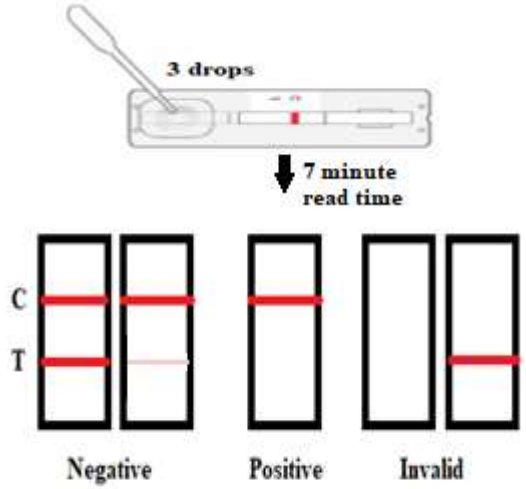
Specimen collection and Preparation

Specimen collection: 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 particles should be centrifuged, filtered, or allowed to settle to obtain a clear urine specimen.
Specimen storage: Urine specimens may be stored at 2-8ºC for up to 48 hours. For long-term storage, specimens may be frozen and stored below -20ºC.

Directions for Use

Allow test cassette and urine specimen to come to room temperature (15-30ºC/59-86ºF) prior to testing.

1. Remove the test cassette and transfer pipette from the foil pouch. Lay the device on a flat and dry surface.
2. Use the transfer pipette supplied to transfer the urine by depressing the bulb of the pipette.
3. Hold the pipette in a vertical position over the cassette. Add 3 drops, drop by drop, roughly 1-2 seconds apart to the sample well.
4. Begin a timer for 7 minutes, at the 7-minute mark read the result. **DO NOT INTERPRET RESULTS AFTER 10 MINUTES.**



Interpretation of Results

Negative: Two lines appear, one in the test region (T) and one in the control region (C). ***Note:** A line in the test region no matter how faint indicates a negative result. This indicates that the Norfentanyl concentration is below the cutoff (60 ng/ml)
Positive: One red line appears in the control region (C). No line appears in the test region. This result indicates that the Norfentanyl concentration is above the cutoff (60 ng/ml)
Invalid: Control line fails to appear. Insufficient specimen volume or incorrect procedural techniques are the mostly likely cause. Check the expiration date. Repeat the test with a new cassette. If the problem persists, discontinue using the kit and contact the manufacturer.

Quality Control

A procedural control is included in the test. A red line appearing in the control region (C) is the internal procedural control. External controls are not supplied with the kit; however, it is recommended that a positive control and a negative control (do not use water) be tested as good laboratory practice to confirm the test procedure and to verify proper test performance.

Limitations of the assay

1. **This assay provides only a preliminary analytical test result. A more specific alternate chemical method must be used to obtain a confirmed analytical result. Gas chromatography/mass spectrometry (GC/MS) is the preferred confirmatory method. Clinical consideration and professional judgement should be applied to any drug of abuse test result, particularly when preliminary positive results are used.**
2. Adulterants, such as bleach or dilute urine specimens may produce erroneous results regardless of the method used.
3. A positive result indicates the presence of the drug or its metabolites but does not indicate the level of intoxication, administration route, or concentration in urine.
4. A negative result does not indicate drug-free urine. A negative result can be obtained when drug is present but below the cut-off.
5. The test does not distinguish between drugs of abuse and certain medications.

Performance Characteristics

Reproducibility

Drug-free urine was spiked with Norfentanyl to ±50% and 100% of the cut-off levels. The results are summarized below.

Norfentanyl concentration	Total number of determinations	Result	Precision
No drug present	20	20 negative	>99%
30 ng/ml	20	20 negative	>99%
90 ng/ml	20	20 positive	>99%
120 ng/ml	20	20 positive	>99%

Effects of Urinary pH

The pH of drug-free urine was adjusted to pH ranges of 5,6,7,8 and 9 and spiked with Norfentanyl ±50% of the cutoff levels. The results demonstrated that varying ranges of pH do not interfere with the performance of the test.

Effects of Specific gravity

Drug-free urine samples with specific gravities of 1.005, 1.01, 1.02, 1.03, and 1.04 were spiked with Norfentanyl ±50% of the cutoff levels. The results demonstrated that varying ranges of specific gravity do not interfere with the performance of the test.

Cross-reactivity between Fentanyl and derivatives

The following table lists the concentration of compounds that were detected positive in urine by the test at a read time of 7 minutes.

Drug	Concentration
Fentanyl	180 ng/ml
Furanyl Fentanyl	>10,000 ng/ml
Acryl Fentanyl	180 ng/ml
Butyryl Fentanyl	340 ng/ml
Acetyl Norfentanyl	600 ng/ml
Butyryl Norfentanyl	100 ng/ml
Isobutyryl Norfentanyl	90 ng/ml