First Sign ®

One-Step Combo (Urine/Serum) Pregnancy Test Cassette Package Insert

For Professional and *In Vitro* Diagnostic Use Only

A Qualitative Test for Detection of Human Chorionic Gonadotropin (hCG) in Human Urine or Serum for Early Detection of Pregnancy

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INTENDED USE

The **First Sign® One-Step Combo (Urine/Serum) Pregnancy Test** is a rapid chromatographic immunoassay designed for the qualitative determination of human chorionic gonadotropin (hCG) in urine or serum for early detection of pregnancy.

SUMMARY AND EXPLANATION

Human chorionic gonadotropin (hCG) is a glycoprotein hormone produced by the developing placenta shortly after fertilization. During normal pregnancy, hCG can be detected in both urine and serum as early as 7 to 10 days following conception. hCG levels continue to rise very rapidly, frequently exceeding 100mIU/mL by the first missed menstrual period, and peaking in the 100,000 to 200,000mIU/mL range about 10 to 12 weeks into pregnancy. The appearance of hCG in both urine and serum soon after conception, and its subsequent rapid rise in concentration during early gestational growth, make it an excellent marker for the early detection of pregnancy.

PRINCIPLE

The **First Sign® One-Step Combo (Urine/Serum) Pregnancy Test** is a rapid test that qualitatively detects the presence of hCG in urine or serum specimen at the sensitivity of 25mIU/mL or above. The method employs a combination of monoclonal and polyclonal antibodies to selectively detect elevated levels of hCG in urine or serum. At the level of claimed sensitivity, the **First Sign® One-Step Combo (Urine/Serum) Pregnancy Test** shows no cross-reactivity interference from the structurally related glycoprotein hormones hFSH, hLH and hTSH at high physiological levels. The assay is conducted by adding urine or serum specimen to the specimen well of the test device and observing the formation of colored lines. The specimen migrates via capillary action along the membrane to react with the colored conjugate.

Positive specimens react with the specific antibody-hCG-colored conjugate to form a colored line at the test line region of the membrane. Absence of this colored line suggests a negative result. To serve as a procedural control, a colored line will always appear at the control line region if the test has been performed properly.

REAGENTS AND MATERIALS

Ingredients: contains a combination of mouse monoclonal antibodies and polyclonal antibodies (sheep or goat) directed against human hCG.
Mouse monoclonal antibodies and polyclonal antibody on colloidal gold particle.

Material Provided:
Each **First Sign® One-Step Combo (Urine/Serum) Pregnancy Test** cassette individually sealed in a foil pouch.
Each pouch contains:
1. One **First Sign® One-Step Combo (Urine/Serum) Pregnancy Test** cassette
2. One disposable specimen dropper
3. Desiccant

Material Required But Not Provided:
1. Timer
2. Sample container
3. Disposable gloves

No other equipment or reagents are needed.

STORAGE AND STABILITY

Store test device at 2°C to 30°C (36°F to 86°F). The test device is stable until the date imprinted on the pouch label.

PRECAUTION

1. For professional and *in vitro* diagnostic use only.
2. Do not use the test beyond the expiration date indicated on the pouch label.
3. The test device should remain in the sealed pouch until use.
4. All specimens should be considered potentially hazardous and handled in the same manner as an infectious agent.
5. The test device should be discarded in a proper biohazard container after testing.
6. Read directions for use carefully before performing this test.

SPECIMEN COLLECTION AND PREPARATION

Urine Assay:
A urine specimen must be collected in a clean and dry container. A first morning urine specimen is preferred since it generally contains the highest concentration of hCG; however, urine specimens collected at any time of the day may be used. Urine specimens exhibiting visible precipitates should be centrifuged, filtered, or allowed to settle to obtain a clear specimen for testing.

Serum Assay:
Blood should be collected aseptically into a clean tube without anticoagulants. Separate the serum from blood as soon as possible to avoid hemolysis. Use clear non-hemolyzed specimens when possible.

SPECIMEN STORAGE

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

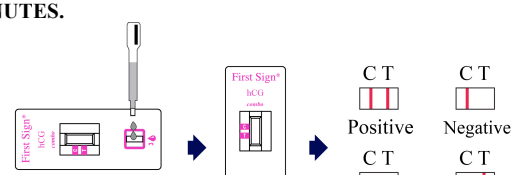
DIRECTIONS FOR USE

Allow the test cassette, urine or serum specimen and/or controls to equilibrate to room temperature (15°C to 30°C) prior to testing.

1. Remove the test cassette from the sealed pouch and use it as soon as possible.
2. Place the test cassette on a clean and level surface. Hold the dropper vertically and transfer 3 full drops of urine or serum (approx. 100µl) to the specimen well of the test cassette, and then start the timer. Avoid trapping air bubbles in the specimen well. See the illustration below.
3. Wait for the pink-purple line(s) to appear**. Read the result at 3 minutes when testing a urine specimen**, **or at 5 minutes when**

testing a serum specimen. It is important that the background is clear before the result is read.

Note: A low hCG concentration might result in a weak line appearing in the test region (T) after an extended period of time; therefore, **DO NOT INTERPRET RESULT AFTER 10 MINUTES.**



INTERPRETATION OF RESULTS

1. **Negative:**
   Only one pink-purple colored band appears in the control area (C).

2. **Positive:**
   In addition to the control band a clearly distinguishable pink-purple colored band also appears in the test area (T).

3. **Invalid:**
   As long as there is no distinct pink-purple colored band visible in control area, then the test is invalid. It is recommended that in this case the test be repeated or fresh specimen be obtained and tested 48 hours later.

QUALITY CONTROL

Built in Quality Control Features:
After addition of the sample, these colored bands migrate along the membrane at the leading edge of the dye conjugate and are "removed" from the test device completely.

When the test is complete, you will see a pink-purple colored band in the "C" area of the test device on negative samples and a pink-purple colored band in the "T" and "C" area on positive samples. The appearance of the CONTROL band indicates that the test device is performing properly and serves as procedural control.

PERFORMANCE CHARACTERISTICS

Negative results are expected in healthy non-pregnant women. Healthy pregnant women have hCG present in their urine and serum specimens. The amount of hCG will vary greatly with gestational age and between individuals.

The **First Sign® One-Step Combo (Urine/Serum) Pregnancy Test** device has a sensitivity of 25mIU/mL, and is capable of detecting pregnancy as early as 1 day after the first missed mensis.

Sensitivity:
The **First Sign® One-Step Combo (Urine/Serum) Pregnancy Test** is a rapid test used to detect the presence of hCG in urine or serum qualitatively at concentrations of 25mIU/mL or greater.

Accuracy:
Test accuracy was assessed by comparing a commercial available pregnancy test (Acon hCG Test). 102 urine and serum samples were tested. 45 urine and serum samples were negative by both methods. 57 urine and serum samples were positive by both methods. **First Sign® One-Step Combo (Urine/Serum) Pregnancy Test** showed 100% agreement with the predicate device: Acon hCG Test.

Specificity:
Potentially interfering substances were added to urine and serum which had hCG levels of 0 and 25mIU/mL. In all case, no interference with the expected result was seen with the **First Sign® One-Step Combo (Urine/Serum) Pregnancy Test**.

Acetaminophen	20mg/dL
Acetysalicylic Acid	20mg/dL
Ascorbic Acid	20mg/dL
Atropine	20mg/dL
Caffeine	20mg/dL
Gentesic Acid	20mg/dL
Glucose	2mg/dL
Hemoglobin	1mg/mL
Ampicillin	20mg/dL
Tetracycline	20mg/dL
Bilirubin (urine)	2mg/dL
Bilirubin (serum)	40mg/dL
Triglycerides (serum)	1200mg/dL
Protein (urine)	500mg/dL

The following hormones were tested for cross reactivity and did not affect the performance of the **First Sign® One-Step Combo (Urine/Serum) Pregnancy Test.**

hTSH	1000μIU/mL	WHO 68/38
hLH	500mIU/mL	WHO 2nd IRP HMG
hFSH	2000mIU/mL	WHO 2nd IRP HMB

LIMITATION

1. Occasionally specimens containing less than 25mIU/mL for urine also yield positive results.
2. In addition to pregnancy, hCG has been found in patients with both gestational and non-gestational trophoblastic disease. Since the hCG has been found in patients with both gestational and non-gestational neoplasms is similar to that found pregnancy, these conditions, which include choriocarcinoma and hydatichiform mole, should be ruled out before a diagnosis of pregnancy is reached.
3. A normal pregnancy can not be distinguished from an ectopic pregnancy based on hCG levels alone. Also, spontaneous miscarriage may cause confusion in interpreting test results.
4. A very early pregnancy containing an extremely low concentration of hCG can give a negative result. In this case, another specimen should be obtained at least 48 hours later and tested.
5. hCG levels may remain detectable for several weeks after normal delivery, delivery by cesarean section, spontaneous abortion or therapeutic abortion.
6. In case where very high levels of hCG are present (>500,000mIU/mL), a false negative result can occur due to a "Prozone" effect. If pregnancy is still suspected, simply dilute specimen 1:1 with deionized water and retest.
7. If a urine sample is too dilute (i.e. low specific gravity), it may not contain a representative level of hCG. If pregnancy is still suspected, a first morning urine sample should be obtained from the patient in 24 to 48 hours and retested.
8. As is true with any diagnostic procedure, the user should evaluate data obtained by the use of this kit in light of other clinical information and consult to with physicians for the final diagnosis of pregnancy before making any decision of medical relevance.

REFERENCE

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