Institution _____ Procedure No. _____

Procedure	SAS™ Serum/Urine hCG

Prepared by	Date Adopted	Supercedes Procedure #

Review Date	Revision Date	Signature

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PRINCIPLE OF THE TEST

SAS™ Serum/Urine hCG is a rapid qualitative test to detect the presence of hCG in serum or urine. The test utilizes a combination of monoclonal and polyclonal antibody reagents to selectively detect elevated levels of hCG in serum or urine. The assay is conducted by the addition of a serum or urine specimen into the test device sample well and observing for the formation of colored lines. The specimen migrates via capillary action along the membrane and reacts with the colored conjugate. A positive specimen reacts with the hCG-specific antibody colored conjugate and forms a colored line in the T (test) window. Absence of this colored line suggests a negative result. To serve as a control for the procedure, a colored line in the C (control) window will always appear regardless of the presence or absence of hCG.

REAGENTS

Test device containing monoclonal mouse-hCG colored conjugate and hCG antibody coated on a membrane.

PRECAUTIONS

1. For *In-Vitro* diagnostic use only.
2. The test device should be discarded in a proper biohazard container after testing.
3. Do not use kit beyond the expiration date.
4. The test device should remain in the sealed pouch until ready for use.

STORAGE AND STABILITY

The test kit is to be stored at room temperature (15° - 30°C) for the duration of the shelf life. The test device must remain "sealed" in the pouch until ready for use.

SPECIMEN COLLECTION AND PREPARATION

Urine - The urine specimen must be collected into a clean, dry container, either plastic or glass. Specimens collected at random may be used; however, the first morning urine generally contains the highest concentration of hormone. A urine sample exhibiting visible precipitates should be filtered, centrifuged, or allowed to settle (obtaining clear aliquots) before testing.

Serum - Blood should be collected aseptically into a clean tube without anticoagulants. Allow clot to form by leaving the tube for 20 to 30 minutes at room temperature. Centrifuge to acquire a clear specimen. If serum shows cloudiness or is highly viscous, it should be diluted with equal parts of saline before testing.

Specimen Storage – Specimens may be refrigerated (2° - 8°C) and stored up to 72 hours prior to assay. If specimens are refrigerated, they must be equilibrated to room temperature (15º - 30ºC) before testing. Serum specimens can be frozen at -20°C for 3 months. Frozen specimens must be thawed and mixed before testing.

PROCEDURE

Materials Provided

1. Test device containing monoclonal mouse-hCG colored conjugate and hCG antibody coated on a membrane.
2. Disposable specimen dropper.

Materials Required But Not Provided

Specimen collection container

Directions For Use

The pouch must be at room temperature before opening to avoid condensation of moisture on the membrane. Allow specimen and/or controls to reach room temperature prior to testing.

1. Remove the test device from the protective pouch and place it on a flat surface. Label the device with patient or control identifications.
2. Dispense 4 drops (approximately 0.15 mL) of specimen into the round sample well (see illustration). Wait for colored lines to appear.

3. Read results after 4 minutes and no later than 15 minutes. Positive results may be observed in as short as 30 seconds depending on the concentration of hCG. READ UNDER DIRECT LIGHT TO AVOID INTERFERENCE OF SHADOWS IN THE T AND C WINDOWS.

INTERPRETATION OF RESULTS

Negative Results
The test is negative if a colored line appears only in the C (control) window.

Positive Results
The test is positive if one colored line appears in the T (test) window and one colored line appears in the C (control) window. Any colored line in the T (test) window should be considered positive. Colored lines may be lighter or darker than each other.

Invalid Results
The test is invalid if no colored line appears in the C (control) window even if a colored line appears in the T (test) window.
Serum - If no colored line appears in the C (control) window or the migration of specimen is slow or incomplete, add 1 to 2 drops of deionized water or saline into the sample well and wait an additional 4 minutes. If a colored line still does not appear in the C (control) window, the serum could be too viscous. Dilute the serum 1:1 with saline or deionized water and repeat the test using another device.
Urine - If no colored line appears in the C (control) window, add 1 to 2 additional drops of urine and wait an additional 4 minutes. If a colored line still does not appear in the C (control) window, the test is invalid and should be repeated using another device.
Colored lines that appear after 15 minutes are not diagnostic and should be ignored.

QUALITY CONTROL
Internal Controls
The appearance of a Control Line in the C region of the device is a positive procedural control. Correct procedural technique, specimen flow and device performance is confirmed when a colored line appears in the C (control) area of the membrane. If the colored line fails to appear in the C (control) area, the test result is invalid.
A clear background is an internal negative procedural control. The background color should be white to light pink and should not interfere with the reading of the test result. If a more intensely red background color appears, it may interfere with the ability to read the test result, therefore the test should be repeated.

External Controls
Urine controls should be used when testing urine. Serum controls should be used when testing serum. Negative and positive controls for hCG should be tested according to federal, state, and local authorities. Quality control should be performed on each lot received. SAS™ controls should be utilized with the SAS™ test kit to ensure proper Q/C testing.

LIMITATIONS OF THE PROCEDURE

1. False negative results may occur when levels of hCG are below the sensitivity level of the test. When pregnancy is still suspected, a fresh serum or a first morning urine specimen should be collected 48 hours later and tested.
2. Elevated levels of hCG may be found in trophoblastic disease, choriocarcinoma, and embryonal cell carcinoma. Islet cell tumors may also produce hCG as well as other carcinomas.[4]
3. Detectable levels of hCG may remain several weeks following normal pregnancy, delivery by caesarean section, spontaneous or therapeutic abortion.[5]
4. Ectopic pregnancies may produce very low levels of hCG.[6] If this condition is suspected, further testing using a quantitative test may be desirable.
5. Approximately one third of all conceptions end in natural termination.[7] This may produce positive results when testing early in the pregnancy, followed by negative results after the natural termination. Low positive results may be confirmed by retesting with a fresh serum or first morning urine specimen collected 48 hours later.
6. This test provides a presumptive diagnosis for pregnancy. Physicians should evaluate all clinical and laboratory findings before making a definitive diagnosis.

EXPECTED VALUES

Negative results are expected in healthy non-pregnant women and healthy men. Healthy pregnant women have hCG present. The amount will vary with gestational age and between patients. SAS™ Serum/Urine hCG can detect hCG levels as low as 25 mIU/mL in serum or urine.

PERFORMANCE CHARACTERISTICS

Accuracy by Comparison

A total of 284 blind clinical samples from suspected pregnant women were studied by different clinics. Samples were assayed with SAS™ Serum/Urine hCG and another commercially available serum & urine test according to assay procedure. Both methods showed 26 positive and 82 negative results in serum testing and 77 positive and 99 negative results in urine testing. The results demonstrated a 100% overall accuracy of SAS™ Serum/Urine hCG compared to the other commercially available test.

Sensitivity & Specificity

SAS™ Serum/Urine hCG detects hCG concentrations of 25 mIU/mL and greater in serum and urine. It has been standardized to World Health Organization Second International Standard (61/6). The addition of LH (300 mIU/mL), FSH (1000 mIU/mL), and TSH (1000 µIU/mL) to negative (0 mIU/mL hCG) and positive (25 mIU/mL hCG) serum/urine showed no cross-reactivity.

Interfering Substances

The following potentially interfering substances were added to negative (0 mIU/mL hCG) and positive (25 mIU/mL hCG) serum/urine samples.

Acetaminophen	20 mg/dL
Acetylsalicylic Acid	20 mg/dL
Ascorbic Acid	20 mg/dL
Atropine	20 mg/dL
Caffeine	20 mg/dL
Gentisic Acid	20 mg/dL
Glucose	2 g/dL
Hemoglobin	1 mg/dL

None of the substances at the concentration tested interfered in the assay.

REFERENCES

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