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For the rapid detection of human chronic gonadotropin (hCG) in human urine specimens
For *in vitro* diagnostic use

INTENDED USE

The DRG® International hCG Urine Rapid Test is intended for detecting the presence of hCG in urine specimens in a qualitative format sensitive to 20 mIU/ml. This test is for in vitro screenning use in obtaining a visual qualitative result for the early detection of pregnancy.

INTRODUCTION

Human chronic gonadotropin (hCG) is a glycoprotein hormone secreted by the developing placenta shortly after fertilization. The hCG appears soon after conception and only during early stages of pregnancy. The DRG® hCG Rapid Test is a qualitative, two-site sandwich immunoassay for the determination of hCG antigen. The assay employs a combination of hCG monoclonal antibodies to selectively detect elevated levels of hCG in human urine specimens. The immunological specificity of the test kit virtually eliminates cross reactivity with the structurally related glycoprotein hormones hFSH, hLH and hTSH at physiological levels.

PRINCIPLE

The DRG® hCG Rapid Test is a chromatographic immunoassay (CIA) for the rapid qualitative determination of hCG in human urine specimens. The membrane is precoated with hCG specific antibodies on the test region. During the test, the specimen is allowed to react with the hCG monoclonal antibody-colloid gold conjugate pre-dried on the test strip. The mixture then moves upward on the membrane chromatographically by capillary action. For a positive specimen, the conjugate binds to the hCG forming an antibody-antigen complex. This complex binds to hCG antibody as a capture regents on the test region and produces a colored band when hCG concentration is equal to or greater than 20 mIU/ml. Absence of this colored band in the test region suggests a negative result. To serve as a procedural control, a colored band in the control region will always appeared regardless the presence of hCG.

REAGENTS AND MATERIALS SUPPLILED

Test device containing sample reacton unit, pink colored colloidal gold conjugate unit, and a chromatographic membrane unit.
Test instruction for the DRG® hCG Urine Rapid Test.

MATERIALS REQUIRED BUT NOT PROVIDED

Urine sample collection container.
Timer or clock with setting of 5 minutes or more.

STORAGE AND STABILITY

The DRG® hCG Urine Rapid Test can be refrigerated or stored at room temperature (2-28ºC) in a sealed pouch. Avoid freezing.

PRECAUTIONS

For professional use only.
Avoid cross contamination of urine samples by using a new sample collection container for each urine sample.

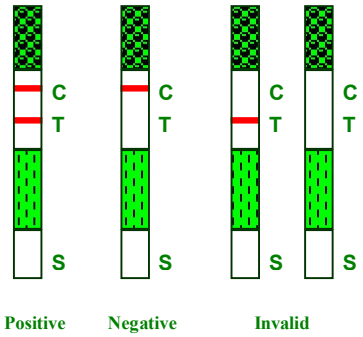
Urine specimens may be potentially infectious. Avoid contact with skin by wearing gloves and proper laboratory attire. Properly handle and dispose of all used test devices in an approved biohazard container.
Test device should remain sealed until ready for use.
Do not use the DRG® hCG Urine Rapid Test devices beyond the expiration date printed on the outside of the foil pouch.

SPECIMEN COLLECTION
Collect and store specimens following standard clinical procedures.
The urine specimen must be collected in a clean, dry container of either plastic or glass and without preservative.
No centrifugation or filtration of urine is required.
Specimens collected at any time may be used, the first morning urine generally contains the highest concentration of hormone.
If specimens cannot be tested after collection, they should be refrigerated at 2-8ºC.

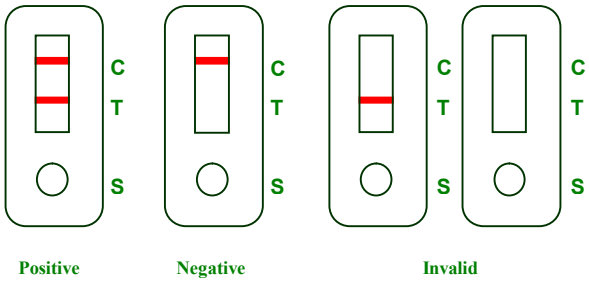
ASSAY PROCEDURE
Do not open foil pouch until ready to begin testing. Refrigerated test devices should be allowed to come to room temperature (15°- 28°C) before opening the pouch.
Remove the test device from the sealed foil pouch by tearing along the notch.
- FOR TEST STRIP: Immerse the strips in urine with the arrow end pointing towards the urine. Do not introduce urine above maximum level line as indicated by the arrows.
- FOR TEST CARD: Draw the urine sample up the pipette and dispense 4 drops (i.e., approximately 0.2 ml) into the sample well of the test device.
Read the DRG® hCG Rapid Test result after 5 minutes.
IMPORTANT: In order to prevent an incorrect reading, do not read the test results after 10 minutes. If the test is read after 10 minutes, the intensity of the colored lines may change or a new line may appear. To avoid confusion, discard all test devices after interpreting results.

INTERPRETATION OF RESULTS
Positive: Presence of two colored bands, one in the control region (C) and another in the test region (T).
Negative: Presence of a colored band in the control region (C) with no band in the test region (T).
Invalid: If no bands appear after 10 minute or if a band appears only in the test region, the result is invalid. If invalid, the protocol may not have been performed correctly or the test has deteriorated over time. The assay should be repeated using a new test device.
Note: Do not interpret result after 10 minutes.

TEST STRIPS



TEST CARDS



QUALITY CONTROL

The DRG® hCG Urine Rapid Test devices have a built-in process Control Region (C). A pink band should always appear in this region, regardless of the presence of any urinary hCG antigen. This pink process Control band verifies: 1) that sufficient urine volume was added, 2) that proper flow is obtained, and 3) that the reagents were working. If this band is missing, the test was not performed correctly or failed to function correctly. Such a test is inconclusive and the specimen should be repeated using a new test device.

Good laboratory practice requires use of control materials to ensure proper test device performance and reliability. The DRG® hCG Urine Rapid Test has been standardized to World Health Organization First International Reference Preparation (WHO, IRP 75/537). Quality control standards are available for the validation of device functionality from commercial sources such as Sigma. Negative and positive controls containing hCG at various concentrations are available commercially. When testing the quality control standards, use the same assay procedure as with a urine sample. It is recommended that control specimens be used for each new lot of kits.

AFTER TESTING

Urine specimens may be potentially infectious. Avoid contact with skin by wearing gloves and proper laboratory attire. Properly handle and dispose of all test devices in an approved biohazard container. Residual urine should be disposed of in a medically approve manner after the completion of all testing.

PERFORMANCE CHARACTERISTICS

Sensitivity

The DRG® hCG Urine Rapid Test has been designed for the detection of hCG in urine at the detection sensitivity of 20 mIU/ml as indicated by the appearance of a colored band on the test region of the test device. Additionally, samples containing less than 20 mIU/ml may also produce a faint positive result. The following sensitivity studies were performed. A hCG standard panel of hCG was prepared by spiking fresh normal human pooled urine specimens with hCG standard (calibrated according to WHO 1st IRP) to concentrations of 0, 5, 10, 15, 20, 25, 50 and 100 mIU/ml. In all, 50 test strip and 50 test card devices per standard level were tested in this procedure (see Table 1 and Table 2).

Table 1: The result of the sensitivity study for the Test Strip

DRG®	HCG Concentration(mIU/ml)							
	0	5	10	15	20	25	50	100
Test No.	50	50	50	50	50	50	50	50
Positive	0	0	23	39	50	50	50	50
Negative	50	50	27	11	0	0	0	0

Table 2: The result of the sensitivity study for the Test Card

DRG®	HCG Concentration(mIU/ml)							
	0	5	10	15	20	25	50	100
Test No.	50	50	50	50	50	50	50	50
Positive	0	0	26	41	50	50	50	50
Negative	50	50	24	9	0	0	0	0

Summary of Sensitivity Results

Concentrations of hCG equal to or lower than 5 mIU/ml were identified as negative for all samples. Concentrations of hCG equal to or higher than 20 mIU/ml were identified as positive results for all samples. Therefore, the sensitivity of the DRG® hCG Rapid Test was determined to be 20 mIU/ml of hCG for the DRG® hCG Urine Rapid Test devices.

Specificity

HOMOLOGOUS HORMONE TESTING

The specificity of the DRG® hCG Urine Rapid Test devices was tested for compounds related or not associated with hCG as prepared in normal human urine. The following compounds were found not to impact negative results when tested at levels up to the concentrations listed:

Luteinizing Hormone (hLH)	300 mIU/ml
Follicle Stimulating Hormone (hFSH)	1000 mIU/ml
Thyroid Stimulating Hormone (hTSH)	1000 mIU/ml

INTERFERENCE TESTING

The following substances were added to urine which had hCG levels of 0 and 50 mIU/ml. None of the substances at concentration tested interfered in the DRG® hCG Urine Rapid Test.

Acetaminophen	20 mg/dl
Acetylsalicyclic Acid	20 mg/dl
Ampicillin	20 mg/dl
Ascorbic Acid	20 mg/dl
Atropine	20 mg/dl
Caffeine	20 mg/dl
Gentesic Acid	20 mg/dl
Glucose	2,000 mg/dl
Hemoglobin	1 mg/dl
Human Serum Protein	2,000 mg/dl
Tetracycline	20 mg/dl
Uric Acid	10 mg/dl

Precision

WITHIN LOT & INTER LOT REPRODUCIBILITY

Summary of Within-Lot Reproducibility:

Three batches of normal human urine demonstrated to be negative for hCG were spiked with hCG to levels of 0, 10, 20, 50 and 100 mIU/ml. Single lots of the DRG® hCG Urine Rapid Test devices were used to test for reproducibility of results. Five devices in each of three batches (for a total of fifteen devices from one lot) were tested per concentration. Results of within-lot reproducibility analytical studies clearly showed excellent repeatability for all 3 batches of positive and negative urine samples, using one lot of the DRG® hCG Urine Rapid Test devices.

Summary of Inter-Lot Reproducibility:

To test inter-lot reproducibility, normal human urine known to be negative for hCG was spiked with hCG to levels of 0, 10, 20, 50 and 100 mIU/ml. The samples were blinded by dispensing the mixed solutions into letter coded labeled vials, and were used to test three (3) lots of Test devices. Twenty (20) devices per concentration for each lot were tested. The results of these tests clearly demonstrate that there is no appreciable inter-lot variation when testing both positive and negative spiked samples across three (3) different lots of the DRG® hCG Urine Rapid Test devices.

KIT COMPARISONS

DRG® HCG TEST DEVICES VS. SURESTEPTM HCG TEST

Accuracy of the DRG® hCG Urine Rapid Test devices was tested against 150 individual in-house laboratory urine samples, as well as against 252 individual external EIA certified clinical laboratory urine samples. The results have been tabulated below as Table 3 and Table 4.

Table 3:

Combined Results: DRG® hCG Test strip vs. SureStepTM Test

DRG®	SureStepTM		Row Totals
	(+)	(-)	
(+)	161	0	161
(-)	0	241	241
Col. Totals	161	241	402

Table 4:

Combined Results: DRG® hCG Test Card vs. SureStepTM Test

DRG®	SureStepTM		Row Totals
	(+)	(-)	
(+)	161	0	161
(-)	0	241	241
Col. Totals	161	241	402

DRG® HCG TEST DEVICES VS. SURESTEPTM AND AXSYM HCG EIA TEST

By means of an external clinical lab, accuracy of the DRG® hCG Urine Rapid Test Devices was evaluated against 252 individual ELISA-certified clinical urine samples. Positive samples and negative samples (total: 252) were analyzed by the quantitative ELISA method approved by the FDA, then these same samples were tested using the DRG® hCG Urine Rapid Test devices. All results were then compared for equivalence and accuracy versus EIA and SureStepTM results, and the data compiled into Table 5 and Table 6 below.

Table 5: Combined Results: DRG® Test vs. Axsym EIA

DRG®	AxsymTM EIA		Row Totals
	(+)	(-)	
(+)	91	8	99
(-)	0	153	153
Col. Totals	91	161	252

When compared to AxsymTM EIA Test data, the relative sensitivity or percent agreement with the DRG® hCG Urine Rapid Test positive samples with the external clinical study was 91/91 or 100%. Negative samples recovered a relative specificity of agreement of 153/161 or 95.0%. The overall relative accuracy obtained was 244/252 or 96.8%.

Table 6: Combined Results - SureStepTM Test vs. Axsym EIA

SureStepT	AxsymTM EIA	Row

M	(+)	(-)	Totals
(+)	91	8	99
(-)	0	153	153
Col. Totals	91	161	252

When compared to AxsymTM EIA Test data, the relative sensitivity or percent agreement with SureStepTM hCG Test positive samples with the external clinical study was 91/91 or 100%. Negative samples recovered a relative specificity of agreement of 153/161 or 95.0%. The overall relative accuracy obtained was 244/252 or 96.8%.

EQUIVALENCY DRG® hCG Urine Rapid Test

Since the DRG® hCG Urine Rapid Test gave equally identical results versus SureStepTM hCG Test data, we concluded that functional equivalency has been demonstrated.

LIMITATIONS

- If a specimen is too diluted (i.e. low specific gravity), it may not contain representative levels of hCG. If pregnancy is still suspected a first morning urine should be obtained from the patient and tested. HCG concentrations of less than 20 mIU/ml will be detected as negative.
- A number of conditions other than pregnancy including trophoblastic disease, proteinuria, hematuria, choriocarcinoma, ovarian and testicular teratomas cause elevated levels of hCG. These diagnoses should be considered if appropriate to the clinical evidence.
- As with all diagnostic tests, a definitive clinical diagnosis should not be based on the results of a single test, but should only be made by the physician after all clinical and laboratory findings have been evaluated.
- Immunologically interfering substances such as those used in antibody therapy treatments may invalidate the test result.

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