



**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY**

I Background Information:

A 510(k) Number

K262299

B Applicant

Alfa Scientific Designs, Inc.

C Proprietary and Established Names

Instant-View® hCG Combo II Cassette Test

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
JHI	II	21 CFR § 862.1155	CH – Clinical Chemistry

II Submission/Device Overview:

A Purpose for Submission:

New device

B Measurand:

Human chorionic gonadotropin (hCG)

C Type of Test:

Qualitative chromatographic immunoassay

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

The Instant-View® hCG Combo II Test is a chromatographic immunoassay for the qualitative detection of human chorionic gonadotropin (hCG) in urine or serum. It is for use as an aid in the early detection of pregnancy. This test is for health care professional use only and not for self-testing.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

D Special Instrument Requirements:

N/A

IV Device/System Characteristics:

A Device Description:

The Instant-View® hCG Combo II Cassette Test is a professional-use only, qualitative assay for the detection of human chorionic gonadotropin (hCG) in urine (20 mIU/mL) or serum (10 mIU/mL) to aid in the early detection of pregnancy. The kit contains 25 test devices, each sealed together with a dropper pipette in a foil pouch, and 1 professional use package insert.

B Principle of Operation:

This assay is a one-step lateral flow chromatographic immunoassay. The test strip in the device consists of a conjugate pad containing a mouse monoclonal anti-hCG antibody conjugated to colloidal gold, and a nitrocellulose membrane precoated with a rabbit monoclonal anti-human hCG α -subunit (CGA) antibody at the test line (T line) and goat anti-mouse IgG at the control line (C line).

When an adequate amount of specimen is applied to the sample well of the device, hCG in the specimen binds to the anti-hCG antibody gold conjugate in the conjugate pad, forming a complex that migrates along the membrane strip. If the hCG level is at or above 20 mIU/mL in urine or 10 mIU/mL in serum, the complex will bind to the capture antibody pre-coated on the T line, developing a colored band. If the specimen contains no hCG or if the level below the detection limit, the T line will not develop.

The goat anti-mouse antibody pre-coated on the C line will always bind to the gold-antibody conjugate, forms a colored line regardless of the presence or absence of hCG.

V Substantial Equivalence Information:

A Predicate Device Name(s):

Alltest Pregnancy Rapid Combo Test Cassette

B Predicate 510(k) Number(s):

K203272

C Comparison with Predicate(s):

Device & Predicate Device(s):	<u>K262299</u>	<u>K203272</u>
Device Trade Name	Instant-View® hCG Combo II Cassette Test	Alltest Pregnancy Rapid Combo Test Cassette
General Device Characteristic Similarities		
Intended Use/Indications For Use	Rapid qualitative detection of hCG to aid in the early detection of pregnancy	Same
Principle of Measurement	Lateral flow Sandwich Immunochromatographic Assay	Same
Cut-off values	Urine: 20 mIU/mL Serum: 10 mIU/mL	Same
General Device Characteristic Differences		
Read time	Urine: 5 minutes Serum: 5 minutes	Urine: 3 minutes Serum: 5 minutes

VI Standards/Guidance Documents Referenced:

None referenced.

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

Negative female serum or urine specimens were spiked with varying hCG (commercially available and traceable to the 6th WHO international Standard) concentrations. The spiked samples were measured in replicates of 6 each day for 5 days using 3 different lots at 3 different testing sites representative of the intended use environment. Tests were performed by 6 different operators for each sample concentration. The cut-off values of urine and serum are 20 mIU/mL hCG and 10 mIU/mL and, respectively. Results are shown in the following tables.

Urine

hCG Concentration (mIU/mL)	Site 1 Lot 3		Site 2 Lot 1		Site 3 Lot 2		Total result		% Negative	% Positive
	-	+	-	+	-	+	-	+		
0	30	0	30	0	30	0	90	0	100	0
5	30	0	30	0	30	0	90	0	100	0

hCG Concentration (mIU/mL)	Site 1 Lot 3		Site 2 Lot 1		Site 3 Lot 2		Total result		% Negative	% Positive
	-	+	-	+	-	+	-	+		
10	30	0	30	0	30	0	90	0	100	0
15	26	4	27	3	27	3	80	10	88.9	11.1
17.5	2	28	2	28	3	27	7	83	7.8	92.2
20	0	30	0	30	0	30	0	90	0	100
25	0	30	0	30	0	30	0	90	0	100
30	0	30	0	30	0	30	0	90	0	100
50	0	30	0	30	0	30	0	90	0	100

Serum

hCG Concentration (mIU/mL)	Site 1 Lot 3		Site 2 Lot 1		Site 3 Lot 2		Total result		% Negative	% Positive
	-	+	-	+	-	+	-	+		
0	30	0	30	0	30	0	90	0	100	0
2	30	0	30	0	30	0	90	0	100	0
5	30	0	30	0	30	0	90	0	100	0
7	4	26	3	27	4	26	11	79	12.2	87.8
8.5	3	27	2	28	2	28	7	83	7.8	92.2
10	0	30	0	30	0	30	0	90	0	100
12	0	30	0	30	0	30	0	90	0	100
15	0	30	0	30	0	30	0	90	0	100
25	0	30	0	30	0	30	0	90	0	100

2. Linearity:
Not applicable. This is a qualitative test.

3. Analytical Specificity/Interference:

Cross-reactivity: The sponsor performed a study to evaluate potential cross-reactivity from LH (Luteinizing hormone), FSH (Follicle-stimulating hormone), and TSH (Thyroid-stimulating hormone). Negative and positive samples (10 and 20 mIU/mL hCG for urine, 5 and 10 mIU/mL hCG for serum) were spiked with various concentrations of LH, FSH, and TSH. Samples were tested using 3 different lots by 3 operators. No interference was observed for these samples for the device at LH concentrations up to 500 mIU/mL, FSH concentrations up to 1000 mIU/mL, and TSH concentrations up to 1000 µIU/mL.

Interference: The sponsor performed a study to evaluate potential interference from exogenous compounds. Each interferent was spiked into both hCG negative (10 mIU/mL for urine, 5 mIU/mL for serum) and hCG positive (20 mIU/mL for urine, 10 mIU/mL for serum) samples. Samples were tested using 3 device lots.

The results showed that no interference was observed from substances at the following concentrations for both negative and positive hCG urine and serum samples. Results are shown in the following table.

Interferent	Concentration
Acetaminophen	20 mg/dL
Acetoacetic Acid	2,000 mg/dL
Acetylsalicylic Acid	20 mg/dL
Albumin	2,000 mg/dL
Ampicillin	20 mg/dL
Ascorbic Acid	20 mg/dL
Atropine	20 mg/dL
B-hydroxybutyrate	2,000 mg/dL
Benzoyllecgonine	10 mg/dL
Bilirubin (urine)	2 mg/dL
Bilirubin (serum)	40 mg/dL
Caffeine	20 mg/dL
Cannabinol	10 mg/dL
Codeine	10 mg/dL
EDTA	80 mg/dL
Ephedrine	20 mg/dL
Estriol-17-beta	1.4 mg/dL
Ethanol	1%
Gentisic Acid	20 mg/dL
Glucose	2,000 mg/dL
Hemoglobin	2,000 mg/dL
High-density lipoprotein (serum only)	70 mg/dL
Ketone	20 mg/dL
Methadone	10 mg/dL
Methanol	10%
Phenothiazine	20 mg/dL
Phenylpropanolamine	20 mg/dL
Pregnanediol	1.5 mg/dL
Salicylic Acid	20 mg/dL
Tetracycline	20 mg/dL
Thiophene	20 mg/dL
Total cholesterol (serum only)	250 mg/dL
Triglycerides (serum only)	1,200 mg/dL

Effect of hCG beta-core fragment

To evaluate potential interference by hCG β -core fragment, negative and positive samples (10 and 20 mIU/mL hCG in urine, 5 and 10 mIU/mL hCG in serum;) were spiked with various concentrations of hCG β -core fragment (0 to 2×10^6 pmol/L). These samples were

tested by 3 different operators using 3 different device lots. No interference from β -core fragment was observed at concentrations up to 2.0×10^6 pmol/L for both urine and serum samples.

Effect of urine pH

To evaluate potential interference from changes in urine pH, negative (i.e., 10 mIU/mL hCG) and positive (i.e., 20 mIU/mL hCG) urine samples were tested at pH 4 to 9 by 3 different operators using 3 different device lots. The data shows that there is no interference from pH ranging from 4 to 9 on tested urine samples.

Effect of urine specific gravity

To evaluate potential interference from changes in specific gravity, negative (i.e., 10 mIU/mL hCG) and positive (i.e., 20 mIU/mL hCG) urine samples were tested at density values ranging from 1.000 to 1.035 by 3 different operators using 3 different device lots. The results indicate that the varying of urine density from 1.000 to 1.035 does not impact the performance of the test device.

Hook effect study

To evaluate high dose hook effect, negative urine and serum samples were spiked with hCG concentrations ranging from 500 to 2,000,000 mIU/mL and were tested in replicates of 5 by 3 different operators using 3 different device lots. The results showed no hook effect up to 2,000,000 mIU/mL for both urine and serum samples tested.

4. Detection Limit and Assay Reportable Range

This assay is intended to detect hCG at levels equal to or above 20 mIU/mL in urine or 10 mIU/mL in serum. See section VII. A.1. for precision of the device near the cutoff and section VII. A.6 for verification of the device's performance at the cut-off.

5. Traceability, Stability, Expected Values (Controls, Calibrators, or Methods):

Traceability

The Instant-View® hCG Combo II Cassette Test is traceable to the WHO 6th International Standard (IS) for human Chorionic Gonadotropin (hCG) 18/244.

Stability

To investigate stability of the device, accelerated stability testing, shelf-life determination, transport simulation testing, and sample stability testing were performed, each by 3 different operators. Based on the results from these studies, the sponsor claims a 24-month shelf life for the device and sample stability for at least 3 days when stored at 2-30°C.

6. Assay Cut-Off:

To verify the cut-off value of the Instant-View® hCG Combo II Cassette Test, negative urine and serum specimens were spiked with varying hCG (commercially available and traceable to the 6th WHO International Standard) concentrations. The spiked samples were randomly labeled and tested using 3 different lots of the device by 3 different

operators. Twenty replicates were tested for each sample concentration and each device lot. Results are shown in the following tables.

Urine

hCG Concentration (mIU/mL)	Lot 1		Lot 2		Lot 3		Total result		% Negative	% Positive
	-	+	-	+	-	+	-	+		
0	20	0	20	0	20	0	60	0	100	0
5	20	0	20	0	20	0	60	0	100	0
10	20	0	20	0	20	0	60	0	100	0
15	19	1	18	2	17	3	54	6	90	10
17.5	1	19	1	19	2	18	4	56	6.7	93.3
20	0	20	0	20	0	20	0	60	0	100
25	0	20	0	20	0	20	0	60	0	100
30	0	20	0	20	0	20	0	60	0	100
50	0	20	0	20	0	20	0	60	0	100

Serum

hCG Concentration (mIU/mL)	Lot 1		Lot 2		Lot 3		Total result		% Negative	% Positive
	-	+	-	+	-	+	-	+		
0	20	0	20	0	20	0	60	0	100	0
2	20	0	20	0	20	0	60	0	100	0
5	20	0	20	0	20	0	60	0	100	0
7	3	17	2	18	2	18	7	53	12	88
8.5	2	18	1	19	1	19	4	56	6.7	93.3
10	0	20	0	20	0	20	0	60	0	100
12	0	20	0	20	0	20	0	60	0	100
15	0	20	0	20	0	20	0	60	0	100
25	0	20	0	20	0	20	0	60	0	100

B Comparison Studies:

1. Method Comparison with Predicate Device:

A total of 124 serum samples and 126 urine were collected from 250 women with ages ranging from 18 to 49 years, who presented to 3 clinical sites for pregnancy testing. All subjects were suspected to be pregnant and all subjects confirmed to be pregnant were in the early stages of pregnancy (less than 5 weeks). The samples were masked and randomized prior to testing by 3 different health professionals using the candidate device

and the predicate device (Alltest Pregnancy Rapid Combo Test Cassette). Three different lots of the proposed device were used in the study for both urine and serum samples. A summary of the results is presented in the tables below:

Urine

New Device	Cleared device	+	-
	+	49	0
	-	0	77

Serum

New Device	Cleared device	+	-
	+	54	0
	-	0	70

The data show that the agreement of the Instant-View® hCG Combo II Cassette Test with the predicate device was 100% for both urine and serum samples.

2. Matrix Comparison:

Not applicable; see sections VII. A.1. and VII. B.1. for precision and method comparison studies conducted with urine and serum samples. Performance with both matrices is described in the performance sections above.

C Clinical Studies:

1. Clinical Sensitivity:

Not applicable.

2. Clinical Specificity:

Not applicable.

3. Clinical Cut-Off:

Not applicable.

4. Other Clinical Supportive Data (When 1. and 2. Are Not Applicable):

Not applicable.

D Expected Values/Reference Range:

Not applicable.

VIII Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device.

IX Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.