Laboratory Name:	
Laboratory Address:	
Date of this packet:	
Insert Revision:	05/27/2014

Fisher Scientific Sure-Vue® Serum/Urine hCG (25 mIU/mL) (30 & 50 Tests) Product No. 4581435021, 4581445021 Laboratory Procedure

This procedure is intended to provide a ready outline reference for performance of the assay. These abbreviated directions for use are not intended to replace the complete package insert. **Any modifications to this document are the sole responsibility of the Facility.**

CLIA Complexity: Waived for Urine and Moderate for Serum

1. Intended Use

The **Sure-Vue® Serum/Urine hCG** is a rapid chromatographic immunoassay for the qualitative detection of human chorionic gonadotropin (hCG) in serum or urine to aid in the early detection of pregnancy.

2. Summary

Human chorionic gonadotropin (hCG) is a glycoprotein hormone produced by the developing placenta shortly after fertilization. In normal pregnancy, hCG can be detected in both urine and serum as early as 7 to 10 days after conception.[1-4] hCG levels continue to rise very rapidly, frequently exceeding 100 mIU/mL by the first missed menstrual period,[2-4] and peaking in the 100,000-200,000 mIU/mL range about 10-12 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.

The **Sure-Vue® Serum/Urine hCG** is a rapid test that qualitatively detects the presence of hCG in serum or urine specimen at the sensitivity of 25 mIU/mL. The test utilizes a combination of monoclonal and polyclonal antibodies to selectively detect elevated levels of hCG in serum or urine. At the level of claimed sensitivity, the **Sure-Vue® Serum/Urine hCG** shows no cross-reactivity interference from the structurally related glycoprotein hormones hFSH, hLH and hTSH at high physiological levels.

3. Test Principle

The **Sure-Vue® Serum/Urine hCG** is a rapid chromatographic immunoassay for the qualitative detection of human chorionic gonadotropin (hCG) in serum or urine to aid in the early detection of pregnancy. The test utilizes a combination of antibodies including mouse monoclonal anti-hCG antibodies and goat polyclonal anti-hCG antibodies to selectively detect elevated levels of hCG. The assay is conducted by adding serum or urine 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 colored antibody conjugates and 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.

4. Specimen Collection/Treatment

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.
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.
Specimen Storage:	Serum or urine specimen may be stored at 2-8°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.
Handling Precautions:	All specimens should be considered potentially hazardous and handled in the same manner as an infectious agent.

5. Reagents

A. Reagents and Materials Provided

Sure-Vue® Serum/Urine hCG- Materials Provided for 30 Test Kit and 50 Test Kit	
Component	**Content**
Test Devices	Contain mouse anti-beta hCG antibody conjugated to colloidal gold and goat anti-alpha hCG antibody coated on the membrane.
Disposable Specimen Droppers	
Package Insert	

B. Materials Required But Not Provided

- Specimen Collection Container
- Timer

6. Storage and Stability

Store as packaged in the sealed pouch at 2-30°C. The test device is stable through the expiration date printed on the sealed pouch. The test device must remain in the sealed pouch until use. **DO NOT FREEZE.** Do not use beyond the expiration date.

7. Quality Control

Internal procedural controls are included in the test. A red line appearing in the control region (C) is the internal procedural control. It confirms sufficient specimen volume and correct procedural technique. A clear background is an internal negative background control. If the test is working properly, the background in the result area should be white to light pink and not interfere with the ability to read the test result.

It is recommended that a positive hCG control (containing ≥ 25 mIU/mL hCG in urine or ≥ 25 mIU/mL hCG in serum) and a negative hCG control (containing "0" mIU/mL hCG) be evaluated to verify proper test performance. For urine testing, controls should be tested with each new lot or shipment of

product, with each new operator, monthly as a check on continued storage conditions, or as otherwise required by your laboratory's internal quality system procedures. For serum testing, federal, state, and local guidelines should be followed.

8. Precautions

- For professional *in vitro* diagnostic use only. Do not use after the expiration date.
- The test device should remain in the sealed pouch until use.
- All specimens should be considered potentially hazardous and handled in the same manner as an infectious agent.
- The test device should be discarded in a proper biohazard container after testing.

9. Test Procedure

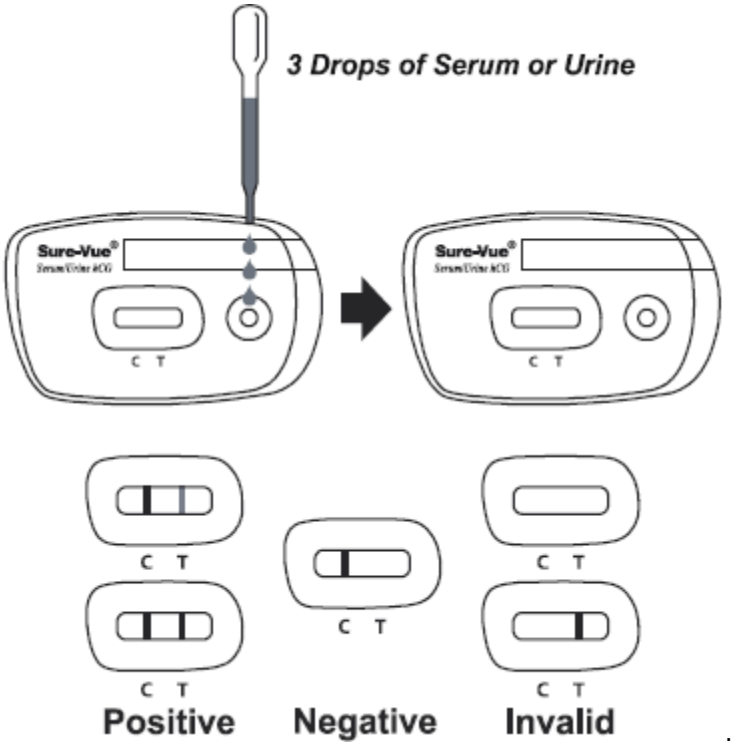
(See the illustration below.)

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

1. Remove the test device from the sealed pouch and use it as soon as possible.

2. Place the test device on a clean and level surface. Hold the dropper vertically and transfer 3 full drops of serum or urine (approx. 100 µL) to the specimen well of the test device, and then start the timer. Avoid trapping air bubbles in the specimen well. See the illustration below.

3. Wait for the red line(s) to appear. **Read the result at 3 minutes when testing a urine specimen, or at 5 minutes when testing a serum specimen. Do not interpret results after the appropriate read time.** It is important that the background is clear before the result is read.



10. Interpretation of Test Results

(Please refer to the illustration above.)

POSITIVE*: Two distinct red lines appear. One line should be in the control region (C) and another line should be in the test region (T).

NEGATIVE: One red line appears in the control region (C). No apparent red or pink line appears in the test region (T).

INVALID: Control line fails to appear. Insufficient specimen volume or incorrect procedural techniques are the most likely reasons for control line failure. Review the procedure and repeat the test with a new test device. If the problem persists, discontinue using the test kit immediately and call 1-866-216-0094 for Technical Assistance.

***NOTE:** The intensity of the red color in the test line region (T) will vary depending on the concentration of hCG present in the specimen. However, neither the quantitative value nor the rate of increase in hCG can be determined by this qualitative test.

11. Limitations

1. Very dilute urine specimens, as indicated by a low specific gravity, may not contain representative levels of hCG. If pregnancy is still suspected, a first morning urine specimen should be collected 48 hours later and tested.
2. False negative results may occur when the levels of hCG are below the sensitivity level of the test. When pregnancy is still suspected, a first morning serum or urine specimen should be collected 48 hours later and tested.
3. Very low levels of hCG (less than 50 mIU/mL) are present in urine and serum specimen shortly after implantation. However, because a significant number of first trimester pregnancies terminate for natural reasons,[5] a test result that is weakly positive should be confirmed by retesting with a first morning serum or urine specimen collected 48 hours later.
4. This test reliably detects intact hCG up to 500,000 mIU/mL. It does not reliably detect hCG degradation products, including free-beta hCG and beta core fragments. Quantitative assays used to detect hCG may detect hCG degradation products and therefore may disagree with the results of this rapid test.
5. A number of conditions other than pregnancy, including trophoblastic disease and certain non-trophoblastic neoplasms including testicular tumors, prostate cancer, breast cancer, and lung cancer, cause elevated levels of hCG.[6,7] Therefore, the presence of hCG in serum or urine specimen should not be used to diagnose pregnancy unless these conditions have been ruled out.
6. As with any assay employing mouse antibodies, the possibility exists for interference by human anti-mouse antibodies (HAMA) in the specimen. Specimens from patients who have received preparations of monoclonal antibodies for diagnosis or therapy may contain HAMA. Such specimens may cause false positive or false negative results.
7. This test provides a presumptive diagnosis for pregnancy. A confirmed pregnancy diagnosis should only be made by a physician after all clinical and laboratory findings have been evaluated.

12. Expected Values

Negative results are expected in healthy non-pregnant women and healthy men. 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 **Sure-Vue® Serum/Urine hCG** has a sensitivity of 25 mIU/mL, and is capable of detecting pregnancy as early as 1 day after the first missed menses.

13. Performance Characteristics

Accuracy

A multi-center clinical evaluation was conducted comparing the results obtained using the **Sure-Vue® Serum/Urine hCG** and another commercially available serum/urine membrane hCG test. The urine study included 159 specimens and both assays identified 88 negative and 71 positive results. The serum study included 73 specimens and both assays identified 51 negative and 21 positive and 1 inconclusive results. The results demonstrated 100% overall agreement (for an accuracy of > 99%) of the **Sure-Vue® Serum/Urine hCG** when compared to the other urine/serum membrane hCG test.

Sensitivity and Specificity
The **Sure-Vue® Serum/Urine hCG** detects hCG at concentrations of 25 mIU/mL or greater. The test has been standardized to the W.H.O. Third International Standard. The addition of LH (300 mIU/mL), FSH (1,000 mIU/mL), and TSH (1,000 µIU/mL) to negative (0 mIU/mL hCG) and positive (25 mIU/ mL hCG) specimens showed no cross-reactivity.

Interfering Substances
The following potentially interfering substances were added to hCG negative and positive specimens.

All substances listed in mg/dL unless otherwise noted.

Acetaminophen	20	Cocaine	10	Ibuprofen	20
Acetone	1,000	Codeine	10	Methadone	10
Acetylsalicylic Acid	20	Cholesterol	500	Methamphetamine	10
Acetoacetic Acid	2,000	Creatine	20	Methanol	10%
Ampicillin	20	Dextramethorphan	20	Morphine	0.6
Ascorbic Acid	20	DMSO	5%	Oxalic Acid	40
Atropine	20	EDTA	80	Phenothiazine	20
Albumin	2,000	Ephedrine	20	Phenylpropanolamine	20
β-Hydroxybutyrate salt	2,000	Ethanol	1%	Pregnanediol	2
Benzoylecgonine	10	Estriol	2	Salicylic Acid	20
Bilirubin	20	Estrone 3-Sulfate	10	Tetracycline	20
Brompheniramine	20	Gentisic Acid	20	Triglycerides	1,200
Caffeine	20	Glucose	2,000	Theophylline	20
Cannabinol	10	Hemoglobin	1,000	Urea	2,000
Clomiphene	100	Heroin	1	Uric acid	20

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

14. References

1. Batzer FR. “Hormonal evaluation of early pregnancy”, *Fertil. Steril.* 1980; 34(1): 1-13
2. Catt KJ, ML Dufau, JL Vaitukaitis “Appearance of hCG in pregnancy plasma following the initiation of implantation of the blastocyte”, *J. Clin. Endocrinol. Metab.* 1975; 40(3): 537-540
3. Braunstein GD, J Rasor, H. Danzer, D Adler, ME Wade “Serum human chorionic gonadotropin levels throughout normal pregnancy”, *Am. J. Obstet. Gynecol.* 1976; 126(6): 678-681
4. Lenton EA, LM Neal, R Sulaiman “Plasma concentration of human chorionic gonadotropin from the time of implantation until the second week of pregnancy,” *Fertil. Steril.* 1982; 37(6): 773-778
5. Steier JA, P Bergsjo, OL Myking “Human chorionic gonadotropin in maternal plasma after induced abortion, spontaneous abortion and removed ectopic pregnancy”, *Obstet. Gynecol.* 1984; 64(3): 391-394
6. Dawood MY, BB Saxena, R Landesman “Human chorionic gonadotropin and its subunits in hydatidiform mole and choriocarcinoma”, *Obstet. Gynecol.* 1977; 50(2): 172-181
7. Braunstein GD, JL Vaitukaitis, PP Carbone, GT Ross “Ectopic production of human chorionic gonadotropin by neoplasms,” *Ann. Intern Med.* 1973; 78(1): 39-45

Test Procedure Approval and Review Sheet

Prepared By:	
Date:	
Supervisor Review:	
Date:	
Laboratory Director or Designee Approval:	
Implementation Date:	
Supersedes Procedure Dated:	
Date Procedure Retired:	

Laboratory Director or Designee	Date Reviewed	Laboratory Director or Designee	Date Reviewed

Fisher Scientific Sure-Vue® Serum/Urine hCG Verification Form

Account Name: _____

Address: _____

_____

Telephone: _____

Fisher Scientific Sure-Vue®
Serum/Urine hCG
Lot #/Exp: _____

Date: _____

Supervisor Signature: _____

Record the results from reference samples below.

Record the Sample #, the **Fisher Scientific Sure-Vue® Serum/Urine hCG** results, Tester's Initials, and any comments. After the **Fisher Scientific Sure-Vue® Serum/Urine hCG** results have been recorded (positive or negative) then record the Expected Results (positive or negative).

Sample #	Expected Results	**Fisher Scientific Sure-Vue® Serum/Urine hCG** Result	Tester's Initials	Comments

Fisher Scientific Sure-Vue® Serum/Urine hCG Verification Form (continued)

Sample #	Expected Results	**Fisher Scientific Sure-Vue® Serum/Urine hCG** Result	Tester's Initials	Comments

Review: _____ Date: _____

Laboratory Director Review and Approval for Clinical Use: _____

Date: _____

Fisher Scientific Sure-Vue® Serum/Urine hCG External Liquid Quality Control Complexity Waived (Urine)

Name of Facility: _____

External Quality Control testing is recommended:

- Controls should be tested with each new lot or shipment of product, with each new operator, monthly or as otherwise required by your laboratory's internal quality system procedures.
- Federal, state, and local guidelines should be followed.

Date	**Fisher Scientific Sure-Vue® Serum/Urine hCG** Kit Lot#/Exp	Date Kit Received	hCG Control Set Lot#/Exp	Positive Control Result	Negative Control Result	Tech's Initials	Comments

Reviewed by:_____ Date:_____

Fisher Scientific Sure-Vue® Serum/Urine hCG External Liquid Quality Control Complexity Moderate (Serum)

Name of Facility: _____

There are two options for complying with CLIA's daily QC requirements for non-waived test systems under Section 493.1256 of the regulations:

- Run two levels of external controls daily before patient testing OR
- Laboratories may develop and implement an IQCP for each non-waived test system.

Alere IQCP Support Documents may be found at http://www.alere.com/IQCP. The following listed conditions are also required as a minimum requirement:

- Controls should be tested with each new lot or shipment of product, with each new operator, monthly or as otherwise required by your laboratory's internal quality system procedures.
- Federal, state, and local guidelines should be followed.

Date	**Fisher Scientific Sure-Vue® Serum/Urine hCG** Lot#/Exp	Date Kit Received	hCG Control Set Lot#/Exp	hCG Positive Control Result	hCG Negative Control Result	Tech's Initials	Comments

Reviewed by: _____ Date: _____

Fisher Scientific Sure-Vue® Serum/Urine hCG Quality Control and Patient Record

Lot Number _____ **Exp. Date** _____

Record the Date, Patient's Name, Patient Test Result, Internal Control Results and the performer's initials.
Positive Internal Control = A red line appearing in the control region (C) is the internal procedural control. Negative Internal Control = A clear background is an internal negative background control.

Date	Patient Name	Patient ID Number	Patient Results	Are the Internal Control Results Invalid or Valid?		Internal Control Results		Comments	Tech
				Invalid	**Valid**	**+**	**-**		

Reviewed by: _____ Date: _____

Fisher Scientific Sure-Vue® Serum/Urine hCG Lot to Lot Comparisons For Non-Waived (Serum) Testing Only

Name of Facility: _____

External Quality Controls are required to test a new lot of reagents.

- When a new shipment or new lot of kit is received
- When required by local, state, and/or federal regulations, accrediting groups, or your lab's Quality Control procedures

	CURRENT Fisher Scientific Sure-Vue® Serum/Urine hCG In-Use Kit				**NEW Kit Fisher Scientific Sure-Vue® Serum/Urine hCG kit**				
Date	**Sure-Vue® Serum/Urine hCG** Kit Lot/ Exp	hCG Control Set Lot/Exp	hCG Positive Ctrl Result	hCG Negative Ctrl Result	**Sure-Vue® Serum/Urine hCG** Kit Lot/ Exp	hCG Control Set Lot/Exp	hCG Positive Ctrl Result	hCG Negative Ctrl Result	User Initials

Reviewed by: _____ Date: _____

Quality Assessment Review Form and Checklist

These forms are used for periodical review of the patient testing process. These should be filed with the quality assessment records.

Quality Assessment Activity	**Comments**	**Date**	**Initials**
Patient Test Management: Evaluate criteria for specimen submission, handling, and rejection; test results requisitions and reporting, accuracy and reliability of reports.			
Quality Control: Assess calibration and control data, reference range verification, errors in reporting results, corrective actions taken with appropriate documentation records.			
Proficiency Testing: Review the effectiveness of corrective actions taken for unsatisfactory performance or failures.			
Comparison of Test Results: Review at least semi-annually comparative results for multiple methods, instruments, or site correlations when more than one procedure exists.			
Relationship of Patient Test Information to Test Results*:* Evaluate patient test reports for accuracy of patient information, test results, and normal ranges. Identify and evaluate results inconsistent with Patient's age, sex, diagnosis, and other test parameters.			
Personnel: Evaluate the effectiveness of policies and procedures for assuring employees competence of testing and reporting test results.			
Communications: Evaluate documented problems and corrective actions that occur between the laboratory and the authorized individual who orders or receives the test result.			
Complaint Investigation: Evaluate documented complaints and corrective actions.			
Quality Assessment Reviews with Staff: Document discussion with Staff regarding identified problems and corrective actions during the QA review.			

Corrective Action Form

Problem/Error	**Corrective Action**

Technologist: _____ Date: _____

Supervisor: _____ Date: _____

Laboratory Director: _____ Date: _____

TEMPERATURE LOG

Equipment: _____

Name of Facility: _____

To be recorded at the beginning of each workday. Temperature Range: _____

Date	°C	Initials	Adjustments	Date	°C	Initials	Adjustments

Tips for Successful Proficiency Testing (PT) Performance

- Strictly follow the PT provider's storage or handling requirement ***before testing PT specimens***.
- Analyze PT specimens ***within the time frame*** provided by the PT provider.
- Contact the PT provider ***promptly*** when specimens are received damaged. You may be able to receive a replacement immediately.
- Avoid clerical error when filling out PT answer sheets. Be sure to ***enter the correct result next to the correct analyte*** on the answer form.
- Remember to identify the instrument or method you are using to perform your PT so you are ***graded among your peer group***.
- Make copies of all answer forms ***before submitting them*** to your PT provider.
- Please contact Technical Support at 877-441-7440 or Lateral.Flow.Support@alere.com for further information on proficiency providers.

Certification of Training

This is to verify that personnel responsible for running the **Fisher Scientific Sure-Vue® Serum/Urine hCG** at _____ have been thoroughly in-serviced on the test and the test procedure. This has included:

- **Review of the package insert**
- **Demonstration of the product assay**
- **Successful performance of the Fisher Scientific Sure-Vue® Serum/Urine hCG and interpretation of results**

Names of the personnel who have been trained with the **Fisher Scientific Sure-Vue® Serum/Urine hCG** and are responsible for reporting patient results:

PRINT NAME	SIGNATURE	DATE

Signature of Laboratory Director(s) responsible for personnel and testing:

_____ _____
Signature Date

_____ _____
Signature Date

_____ _____
Trainer Date

Testing Personnel Training Assessment

Test Method: Fisher Scientific Sure-Vue® Serum/Urine hCG

Procedure	Satisfactory	Unsatisfactory	Not Applicable	Comments / Corrective Actions
Observation of Test Performance:				
Patient Sample Preparation (if applicable)				
Specimen Handling/Processing				
Testing				
Recording/Reporting Results				
Assessment of Test Performance Using Known Samples				
Review of Records:				
Patient/Quality Control Log Sheet Records				
Proficiency Testing Records				
Assessment of Problem Solving Skills				

(Attach all supporting documents)

Evaluator:_____ Date: _____

Employee:_____

Fisher Scientific Sure-Vue® Serum/Urine hCG Quiz

Name:_____

Date: _____

Circle T (True) or F (False) for each Question:

1.	The **Fisher Scientific Sure-Vue® Serum/Urine hCG** test must be refrigerated at 2-8°C.	T	F
2.	The **Fisher Scientific Sure-Vue® Serum/Urine hCG** test pouches may be opened one hour before the test is performed.	T	F
3.	Urine and serum specimens may be refrigerated up to 48 hours prior to testing.	T	F
4.	Four drops of the specimen are added to the **Fisher Scientific Sure-Vue® Serum/Urine hCG** test.	T	F
5.	The **Fisher Scientific Sure-Vue® Serum/Urine hCG** test detection limit is 20 mIU/mL for both serum and urine specimens.	T	F
6.	The **Fisher Scientific Sure-Vue® Serum/Urine hCG** test device should be at room temperature before performing a test.	T	F
7.	A **Fisher Scientific Sure-Vue® Serum/Urine hCG** test device without a red line at the control region, and a red line at the test region, may be reported as positive.	T	F
8.	The **Fisher Scientific Sure-Vue® Serum/Urine hCG** test may be read at three minutes for urine specimens and 5 minutes for serum specimens.	T	F
9.	The **Fisher Scientific Sure-Vue® Serum/Urine hCG** test may be read at 20 minutes.	T	F
10.	If a red line is not visible at the control region, the **Fisher Scientific Sure-Vue® Serum/Urine hCG** test result is invalid.	T	F

Fisher Scientific Sure-Vue® Serum/Urine hCG Quiz Answer Key

	Answer Key	Explanation
1.	F	The **Fisher Scientific Sure-Vue® Serum/Urine hCG** test may be stored refrigerated or at room temperature 2-30°C.
2.	F	The **Fisher Scientific Sure-Vue® Serum/Urine hCG** test devices should remain stored in the pouch until ready to use.
3.	T	The urine and serum specimens may be refrigerated up to 48 hours prior to testing.
4.	F	Three drops of specimen should be added to the sample well using the kit dropper.
5.	F	The detection limit of the **Fisher Scientific Sure-Vue® Serum/Urine hCG** test is 25 mIU/mL for serum and urine specimens.
6.	T	The **Fisher Scientific Sure-Vue® Serum/Urine hCG** test device should be at room temperature prior to testing.
7.	F	If the red control line fails to appear, the test is invalid.
8.	T	The **Fisher Scientific Sure-Vue® Serum/Urine hCG** test may be read at 3 minutes for urine specimens and 5 minutes for serum specimens.
9.	F	The **Fisher Scientific Sure-Vue® Serum/Urine hCG** test results should be read at 3 minutes when testing a urine specimen, or at 5 minutes when testing a serum specimen. Do not interpret results after the appropriate read time.
10.	T	If the red control line fails to appear, the test is invalid.