Diagnostic Automation / Cortez Diagnostics, Inc.
IMMUNO DIAGNOSTICS

CE

OneStep AFP RapiCard™ InstaTest Serum/ Plasma

Cat # 13040-1

IVD 2°C 8°C Σ 1 Test

Sensitivity	**100 %**
Specificity	**98.3%**

INTENDED USE

Cortez Diagnostics, Inc. OneStep AFP test is a colloidal-gold-antibody-complex immunoassay for qualitative determination of human alpha-fetoprotein (AFP) in serum or plasma. It is intended as an aid in the monitoring of patients for disease progression or response to therapy or as an aid in the detection of recurrent or residual disease.

SUMMARY AND EXPLANATION

AFP was first found in the sera of human fetuses in molecular weights ranging from 67,000 to 74,000 daltons; the variations in MW are due to methods of analysis and the different degrees of glycosylation of the protein.

AFP is a single-chain protein showing close sequence homology with serum albumin. It is suggested to play a role in transporting polyunsaturated fatty acids to developing and malignant cells because of its high affinity to these substances. Studies also indicate that AFP may function as an important *in vivo* immunoregulator that acts through T cells.

Synthesis of AFP in the human yolk sac ceases between the tenth and twelfth week of gestation. The major portion of the protein is then produced by fetal hepatocytes. The upper limit for normal sera is about 9 ng/ml, while levels above 175 ng/ml are highly suspicious of hepatocellular carcinoma. While 82% of patients with clinically verified tumors have higher amounts, 98% of patients with metastatic liver disease have below 175 ng/ml. It has been shown that the elevation of serum AFP in benign hepatic diseases is usually transient.

Many studies confirm the presence of AFP in the early stages of fetal open neural tube defect (NTD). In these cases, AFP is thought to leak directly into the amniotic fluid and subsequently enter maternal circulation, thus producing abnormally elevated levels of maternal serum AFP.

TEST PRINCIPLE

The *OneStep* AFP Test is an immunochromatographic assay which utilizes a unique combination of monoclonal antibodies to selectively identify AFP in serum or plasma specimens with a high degree of sensitivity. Elevated levels of AFP are detected in ten minutes or less.

Serum specimen flows through the absorbent area of the reaction strip and migrates along the strip membrane. In the absorbent area, dye-conjugate anti AFP antibody binds to AFP molecules in the specimen forming a colored antibody-antigen complex. Anti-AFP antibody immobilized in the test zone of the membrane captures the antibody-antigen complex causing a visible rose-pink color band, indicating a positive result. In the control zone of the membrane, immobilized reagents capture colored conjugate regardless of the specimen composition. The resulting visible rose-pink color band confirms that the assay is functioning correctly. No line formation in the test zone indicates a negative reading or that the AFP level of the specimen is below the detection sensitivity of the test.

SPECIMEN COLLECTION AND PREPARATION

Collect blood aseptically by venipuncture into a clean tube without anticoagulants. Permit blood to clot for twenty to thirty minutes at room temperature. Centrifuge to obtain clear serum and transfer serum into a clean plastic or glass tube. The test may be performed using human serum or plasma.

If specimens are not immediately tested they should be refrigerated at 2-8° C. For storage periods greater than three days, freezing is recommended (- 20ºC). If specimens are to be shipped, they should be packed in compliance with federal regulations covering the transportation of etiologic agents.

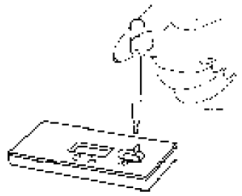
Specimens containing precipitate may yield inconsistent test results. Such specimens must be clarified prior to assaying.

ASSAY PROCEDURE

Bring unopened test components and sample specimens to room temperature prior to testing.

1. Open a foil pouch by tearing along the splice and remove the test cassette and sample dropper.
2. Holding the dropper vertically, add four full drops of sample specimen without air bubbles to the sample well "S" of the test device.
3. Read the result at ten minutes.

IMPORTANT: THE RESULT MUST BE INTERPRETED AT FIVE MINUTES. WAITING MORE THAN FIVE MINUTES MAY CAUSE THE READING TO BE INACCURATE. TO AVOID CONFUSION, DISCARD THE TEST DEVICE AFTER INTERPRETING THE RESULT.



MATERIALS AND COMPONENTS

Materials provided with the test kits

1. Test Cassette:
   An absorbent device with an antibody coated membrane and a pad treated with mouse monoclonal IgG-dye conjugate in a protein matrix containing sodium azide.
2. Sample Dropper:
   A transfer pipette is included with each test device inside the foil pouch.
3. Test Instructions.

Materials required but not provided

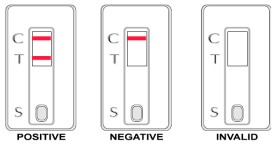
1. Specimen collection container.
2. Centrifuge capable of 1000 x *g* (for centrifuging whole blood specimens).
3. Clock or timer.

Diagnostic Automation/Cortez Diagnostics, Inc.
23961 Craftsman Road, Suite D/E/F, Calabasas, CA 91302 USA Phone: 818.591.3030 Fax 818.591.8383
Email:onestep@rapidtest.com Website: www.rapidtest.com

RESULTS

- **Negative:** One rose-pink color band appears in the Control Zone "C" with no apparent color band in the Test Zone "T". The AFP level of the specimen is below the 20 ng/ml detection cutoff of the test.
- **Positive:** Two rose-pink color bands appear in the result window, one in the Control Zone "C" and one in Test Zone "T." A positive result indicates AFP is present in the sample at or above the 20 ng/ml detection cutoff.
- **Invalid:** If no rose-pink color band is visible in the control zone "C," the test result is invalid. Retest the specimen using a new test device.

Note: There is no meaning attributed to line color intensity or width.



PERFORMANCE CHARACTERISTICS

1. **Sensitivity**

The analytical sensitivity of the *OneStep AFP* Test is 20 ng/ml.

2. **Accuracy**

A study was performed using ninety-five positive and negative serum specimens. Each specimen was assayed with the *OneStep AFP* Test and a commercially available AFP test according to the respective package insert instructions.

Correlation Study

DAI	Commercial Test
+/+	+/-
37	1
-/+	-/-
0	58

Relative Sensitivity: 100 %
Relative Specifity: 98.3 %

The data demonstrates an excellent correlation between the two tests. The clinical significance of the two tests is comparable.

QUALITY CONTROL

An internal procedure control has been incorporated into the test to ensure proper kit performance and reliability.

The use of a control is recommended to verify proper kit performance. Quality control samples should be tested according to quality control requirements established by the testing laboratory.

Use the control in the same manner as a specimen by following the test procedure. The expected results should be obtained when using the control.

LIMITATIONS OF PROCEDURE

1. The test is limited to the detection AFP in serum, plasma, or recalcified plasma.
2. The test is for research use only.
3. Although the test is very accurate in detecting elevated AFP levels, a low incidence of false results may occur.
4. The test is a qualitative screening assay and is not suggested for quantitative APF determination.
5. 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.

PRECAUTION

This kit contains no infectious reagents, however proper precautions should always be taken when handling patient specimens.

1. Preclude any pipetting by mouth.
2. Do not allow smoking or eating where specimen and reagents are being handled.
3. Wear disposable gloves while handling kit reagents or specimens. Wash hands thoroughly afterwards.
4. Avoid splashing or aerosol formation.
5. Clean up spills thoroughly using an appropriate intermediate-to-high level disinfectant.
6. Decontaminate and dispose of all specimens and potentially contaminated materials as if they were infectious.
7. Do not use reagents after the expiration date.
8. For *in vitro* diagnostic use only.

STORAGE AND STABILITY

Store the test kit at 2°-8° C; do not freeze. Prior to use, bring test components to room temperature. The testing device may be stored at room temperature (below 28ºC).

ISO 13485
ISO 9001

Diagnostic Automation/
Cortez Diagnostics, Inc.
23961 Craftsman Road, Suite E/F,
Calabasas, California 91302 USA

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EC REP	CEpartner4U, Esdoornlaan 13, 3951DB Maarn. The Netherlands. www.cepartner4u.eu
Revision B Date: 01-15-2015	