

CA 125 II™

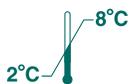
Customer Service

United States: 1-877-4Abbott

International: Call your Abbott Representative

This package insert must be read carefully prior to use. Package insert instructions must be followed accordingly. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions in this package insert.

Key to symbols used

REF	List number	LOT	Lot number
IVD	For <i>In Vitro</i> Diagnostic Use	CAL A	Calibrator (A-F)
	Store at 2-8°C	CONTROL L	Control Low, Medium, High (L, M, H)
	Consult instructions for use.	ASSAY CD-ROM	Assay CD-ROM
SN	Serial Number	REACTION VESSELS	Reaction Vessels
REAGENT LOT	Reagent Lot	SAMPLE CUPS	Sample Cups
	Expiration Date	SEPTUM	Septum
	Legal Manufacturer	REPLACEMENT CAPS	Replacement Caps
EC REP	Authorized Representative		



EC REP ABBOTT
Max-Planck-Ring 2
65205 Wiesbaden
Germany
+49-6122-580

See **REAGENTS** section for a full explanation of symbols used in reagent component naming.

Produced for



WARNING: CA 125 assay values obtained with different assay methods cannot be used interchangeably due to differences in assay methods and reagent specificity. The results reported by the laboratory to the physician must include the identity of the CA 125 assay used. If, in the course of monitoring a patient, the assay method used for determining serial CA 125 levels is changed, additional sequential testing should be carried out. Prior to changing assays, the laboratory MUST confirm baseline values for patients being serially monitored.

NAME

ARCHITECT CA 125 II

INTENDED USE

The ARCHITECT CA 125 II assay is a Chemiluminescent Microparticle Immunoassay (CMIA) for the quantitative determination of OC 125 defined antigen in human serum and plasma on the ARCHITECT *i* System. The ARCHITECT CA 125 II assay is to be used as an aid in monitoring response to therapy for patients with epithelial ovarian cancer. Serial testing for patient CA 125 II assay values should be used in conjunction with other clinical methods used for monitoring ovarian cancer.

SUMMARY AND EXPLANATION OF TEST

CA 125 II assay values are defined by using the OC 125 monoclonal antibody. OC 125 was generated through the hybridization of mouse myeloma cells to spleen cells from a mouse immunized with a human serous cystadenocarcinoma cell line called OVCA 433.¹ ARCHITECT CA 125 II is a second-generation assay for the detection of OC 125 defined antigen. The assay utilizes the OC 125 monoclonal antibody, as the capture antibody coated onto paramagnetic microparticles that bind molecules containing OC 125 defined antigen. These defined antigens are quantified using acridinium-labeled M11 antibody. The OC 125 monoclonal antibody is reactive with repeating OC 125 defined antigen expressed by a high percentage of nonmucinous ovarian carcinomas (serous, endometrioid, clear cell, and undifferentiated histologies) and epithelial ovarian carcinoma cell lines.^{1,2} OC 125 defined antigens were originally detected in normal peritoneal, pleural and pericardial tissues of both fetus and adult. In the fetus, OC 125 defined antigens have been localized in amniotic and umbilical epithelial and Müllerian epithelial tissues. In the adult, localization has been identified in endocervical and endometrial tissues and ovarian inclusion cysts and papillary excrescences. However, OC 125 defined antigens were not detected in fetal ovarian tissue or other normal adult ovarian tissues or benign mucinous ovarian tumors.³ In serum, the OC 125 defined antigens are associated with high molecular weight glycoproteins heterogeneous in size and charge. The structure of the CA 125 molecule, including closely situated repeating epitopes for OC 125 and M11 antibodies has been proposed.⁴

Serum CA 125 II assay values are useful for monitoring the course of disease in patients with invasive epithelial ovarian cancer.⁵ In a review of nine published studies, the overall correlation reported between CA 125 serum levels and the course of the disease was 87%.⁶ Persistently rising CA 125 assay values may be associated with malignant disease and poor response to therapy, whereas decreasing CA 125 assay values may indicate a favorable response to therapy.⁶⁻¹⁴

A second-look, exploratory laparotomy may have been performed previously to assess response to therapy. The benefit has recently come into question because of high morbidity and low sensitivity in detecting residual or recurrent carcinoma.¹⁵ In women with primary epithelial ovarian carcinoma who had undergone first-line therapy and were candidates for diagnostic second-look procedures, a CA 125 assay value greater than or equal to 35 U/mL was found to be indicative of the presence of residual tumor.^{6,9,11,13} However, a CA 125 assay value below 35 U/mL does not indicate the absence of residual ovarian cancer because patients with histopathologic evidence of ovarian carcinoma may have CA 125 assay values within the range of normal individuals.^{7,8}

Elevations of CA 125 assay values have been reported in approximately 1-2% of healthy individuals,^{6,7} and in individuals with nonmalignant conditions such as cirrhosis,^{16,17} hepatitis,^{17,18} endometriosis,¹⁹⁻²⁴ first trimester pregnancy,²⁵⁻²⁷ ovarian cysts,^{3,28} and pelvic inflammatory disease.^{10,25} Elevations of CA 125 assay values during the menstrual cycle have also been reported.^{23,29} Non-ovarian malignancies in which CA 125 assay values have been reported include endocervical,³⁰ liver,¹⁸ pancreatic,^{18,31} lung,¹⁸ colon,^{18,31} stomach,^{18,31} biliary tract,^{18,31} uterine,¹⁷ fallopian tube,³⁰ breast,¹⁸ and endometrial carcinomas.^{30,32} The CA 125 assay is not recommended as a screening procedure to detect cancer in the general population; however, the use of CA 125 assay values as an aid in the management of ovarian cancer patients has been reported.⁷⁻¹⁴

BIOLOGICAL PRINCIPLES OF THE PROCEDURE

The ARCHITECT CA 125 II assay is a two-step immunoassay to determine the presence of OC 125 defined antigen in human serum and plasma, using Chemiluminescent Microparticle Immunoassay (CMIA) technology with flexible assay protocols, referred to as Chemiflex®. In the first step, sample and OC 125 coated paramagnetic microparticles are combined. OC 125 defined antigen present in the sample bind to the OC 125 coated microparticles. After washing, M11 acridinium-labeled conjugate is added in the second step. Pre-Trigger and Trigger Solutions are then added to the reaction mixture; the resulting chemiluminescent reaction is measured as relative light units (RLUs). A direct relationship exists between the amount of OC 125 defined antigen in the sample and the RLUs detected by the ARCHITECT *i* optical system.

For additional information on system and assay technology, refer to the ARCHITECT System Operations Manual, Section 3.

**i* = immunoassay

REAGENTS

Reagent Kit, 100 Tests/500 Tests

NOTE: All kit sizes are not available in some countries, please contact your local distributor.

ARCHITECT CA 125 II Reagent Kit 2K45

- **MICROPARTICLES** 1 or 4 Bottle(s) (6.6 mL for the 100 test bottle/27.0 mL for the 500 test bottle) anti-CA 125 (mouse, monoclonal) coated Microparticles in TRIS buffer with protein (bovine) stabilizers. Preservative: antimicrobial agent.
- **CONJUGATE** 1 or 4 Bottle(s) (5.9 mL for the 100 test bottle/26.3 mL for the 500 test bottle) anti-CA 125 (mouse, monoclonal) acridinium-labeled Conjugate in phosphate buffer with protein (bovine) stabilizers. Minimum concentration: 0.075 µg/mL. Preservative: antimicrobial agent.

Other Reagents

ARCHITECT *i* Pre-Trigger Solution

- **PRE-TRIGGER SOLUTION** Pre-Trigger Solution containing 1.32% (w/v) hydrogen peroxide.

ARCHITECT *i* Trigger Solution

- **TRIGGER SOLUTION** Trigger Solution containing 0.35N sodium hydroxide.

ARCHITECT *i* Wash Buffer

NOTE: Bottle and volume varies based on order.

- **WASH BUFFER** Wash Buffer containing phosphate buffered saline solution. Preservative: antimicrobial agent.

Assay Diluent

ARCHITECT *i* Multi-Assay Manual Diluent (No. 7D82-50)

- **MULTI-ASSAY MANUAL DILUENT** 1 Bottle (100 mL) ARCHITECT *i* Multi-Assay Manual Diluent containing phosphate buffered saline solution. Preservative: antimicrobial agent.

WARNINGS AND PRECAUTIONS

- **IVD For In Vitro Diagnostic Use.**
- Package insert instructions must be followed accordingly. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions in this package insert.

Safety Precautions

- This product requires the handling of human specimens. It is recommended that these reagents and human specimens be handled in accordance with the OSHA Standard on Bloodborne Pathogens³³. Biosafety Level 2³⁴ or other appropriate biosafety practices^{35,36} should be used for materials that contain or are suspected of containing infectious agents.
- The Microparticles and Conjugate contain a mixture of: 5-chloro-2-methyl-4-isothiazolin-3-one and 2-methyl-4-isothiazolin-3-one (3:1) which is a component of ProClin 300® and are classified per applicable European Community (EC) Directives as: Irritant (Xi). The following are the appropriate Risk (R) and Safety (S) phrases.

	R43	May cause sensitization by skin contact.
	S24	Avoid contact with skin.
	S35	This material and its container must be disposed of in a safe way.
	S37	Wear suitable gloves.
	S46	If swallowed, seek medical advice immediately and show this container or label.
- ARCHITECT *i* Trigger Solution contains sodium hydroxide (NaOH) and is classified per applicable European Community (EC) Directives as: Irritant (Xi). The following are the appropriate Risk (R) and Safety (S) phrases.

	R41	Risk of serious damage to eyes.
	S25	Avoid contact with eyes.
	S26	In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.
	S35	This material and its container must be disposed of in a safe way.
	S36/39	Wear suitable protective clothing and eye/face protection.
	S46	If swallowed, seek medical advice immediately and show this container or label.

- Information for European customers: For product not classified as dangerous per European Directive 1999/45/EC - Safety data sheet available for professional user on request.
- For a detailed discussion of safety precautions during system operation, refer to the ARCHITECT System Operations Manual, Section 8.

Handling Precautions

- Do not use reagent kits beyond the expiration date.
- Do not mix reagents from different reagent kits.
- Prior to loading the ARCHITECT CA 125 II Reagent Kit on the system for the first time, the microparticle bottle requires mixing to resuspend microparticles that have settled during shipment. For microparticle mixing instructions, refer to the **PROCEDURE, Assay Procedure** section of this package insert.
- Septa MUST be used to prevent reagent evaporation and contamination and to ensure reagent integrity. Reliability of assay results cannot be guaranteed if septa are not used according to the instructions in this package insert.**
- To avoid contamination, wear clean gloves when placing a septum on an uncapped reagent bottle.**
- Prior to placing the septum on an uncapped reagent bottle, squeeze the septum in half to confirm that the slits are open. If the slits appear sealed, continue to gently squeeze the septum to open the slits.
- Once a septum has been placed on an open reagent bottle, **do not invert the bottle** as this will result in reagent leakage and may compromise assay results.
- Over time, residual liquids may dry on the septum surface. These are typically dried salts, which have no effect on assay efficacy.
- For a detailed discussion of handling precautions during system operation, refer to the ARCHITECT System Operations Manual, Section 7.

Storage Instructions

-  The ARCHITECT CA 125 II Reagent Kit must be stored at 2-8°C in an upright position and may be used immediately after removal from 2-8 °C storage.
- When stored and handled as directed, reagents are stable until the expiration date.
- The ARCHITECT CA 125 II Reagent Kit may be stored on-board the ARCHITECT *i* System for a maximum of 30 days. After 30 days, the reagent kit must be discarded. For information on tracking on-board time, refer to the ARCHITECT System Operations Manual, Section 5.
- Reagents may be stored on or off the ARCHITECT *i* System. If reagents are removed from the system, store them at 2-8°C (with septa and replacement caps) in an upright position. For reagents stored off the system, it is recommended that they be stored in their original trays and boxes to ensure they remain upright. **If any reagent bottle does not remain upright (with a septum installed) while in refrigerated storage off the system, the reagent kit must be discarded.** After reagents are removed from the system, you must initiate a scan to update the on-board stability timer.

Indications of Reagent Deterioration

When a control value is out of the specified range, it may indicate deterioration of the reagents or errors in technique. Associated test results may be invalid and may require retesting. Assay recalibration may be necessary. For troubleshooting information, refer to the ARCHITECT System Operations Manual, Section 10.

INSTRUMENT PROCEDURE

- The ARCHITECT CA 125 II assay file must be installed on the ARCHITECT *i* System from the ARCHITECT *i* Assay CD-ROM Addition A prior to performing the assay. For detailed information on assay file installation and on viewing and editing assay parameters, refer to the ARCHITECT System Operations Manual, Section 2.
- For information on printing assay parameters, refer to the ARCHITECT System Operations Manual, Section 5.
- For a detailed description of system procedures, refer to the ARCHITECT System Operations Manual.

SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS

- Only human serum (including serum collected in separator tubes [SST®]) or plasma (collected in tripotassium EDTA, sodium heparin, or lithium heparin collection tubes) may be used in the ARCHITECT CA 125 II assay. Other anticoagulants have not been validated for use with the ARCHITECT CA 125 II assay. Follow the tube manufacturer's processing instructions for collection tubes.
- When serial specimens are being evaluated, the same type of specimen should be used throughout the study.
- The ARCHITECT *i* System does not provide the capability to verify specimen type. It is the responsibility of the operator to verify that the correct specimen type is used in the ARCHITECT CA 125 II assay.

- Use caution when handling patient specimens to prevent cross contamination. Use of disposable pipettes or pipette tips is recommended.
- Do not use grossly hemolyzed specimens.
- Do not use heat-inactivated specimens.
- Inspect all samples for bubbles. Remove bubbles with an applicator stick prior to analysis. Use a new applicator stick for each sample to prevent cross contamination.
- Serum and plasma specimens should be free of fibrin, red blood cell or other particulate matter.
- Ensure that complete clot formation in serum specimens has taken place prior to centrifugation. Some specimens, especially those from patients receiving anticoagulant or thrombolytic therapy, may exhibit increased clotting time. If the specimen is centrifuged before a complete clot forms, the presence of fibrin may cause erroneous results.
- If testing will be delayed more than 24 hours, remove serum or plasma from the clot, serum separator, or red blood cells. Specimens may be stored for up to 7 days at 2-8°C prior to being tested. If testing will be delayed more than 7 days, serum or plasma should be stored frozen at -20°C or colder.
- Multiple freeze-thaw cycles of specimens should be avoided. Specimens must be mixed THOROUGHLY after thawing, by LOW speed vortexing or by gently inverting, and centrifuged prior to use to remove red blood cells or particulate matter to ensure consistency in the results. Sample from the middle of the tube to avoid any particulate on the top or bottom of the sample.
- Performance has not been established using body fluids other than human serum and plasma.
- Specimens with obvious microbial contamination should not be used.
- Prior to shipment, it is recommended that specimens be removed from the clot, serum separator or red blood cells. When shipped, specimens must be packaged and labeled in compliance with applicable state, federal, and international regulations covering the transport of clinical specimens and infectious substances. Specimens may be shipped on wet or dry ice.

PROCEDURE

Materials Provided:

- 2K45 ARCHITECT CA 125 II Reagent Kit

Materials Required but not Provided:

- ARCHITECT *i* System
- 3K50 ARCHITECT *i* **ASSAY CD-ROM** -US-Addition A
- 3K52 ARCHITECT *i* **ASSAY CD-ROM** -WW (excluding US)-Addition A
- 2K45-01 ARCHITECT CA 125 II Calibrators
- 2K45-10 ARCHITECT CA 125 II Controls
- 7D82-50 ARCHITECT *i* **MULTI-ASSAY MANUAL DILUENT**
- ARCHITECT *i* **PRE-TRIGGER SOLUTION**
- ARCHITECT *i* **TRIGGER SOLUTION**
- ARCHITECT *i* **WASH BUFFER**
- ARCHITECT *i* **REACTION VESSELS**
- ARCHITECT *i* **SAMPLE CUPS**
- ARCHITECT *i* **SEPTUM**
- ARCHITECT *i* **REPLACEMENT CAPS**
- Pipettes or pipette tips (optional) to deliver the volumes specified on the patient or control order screen.
- For information on materials required for maintenance procedures, refer to the ARCHITECT System Operations Manual, Section 9.

Assay Procedure

- Before loading the ARCHITECT CA 125 II Reagent Kit on the system for the first time, the microparticle bottle requires mixing to resuspend microparticles that have settled during shipment:
 - Invert the microparticle bottle 30 times.
 - Visually inspect the bottle to ensure microparticles are resuspended. If microparticles are still adhered to the bottle, continue to invert the bottle until the microparticles have been completely resuspended.
- Once the microparticles have been resuspended, remove and discard the cap. Wearing clean gloves, remove a septum from the bag. Squeeze the septum in half to confirm that the slits are open. Carefully snap the septum onto the top of the bottle.
 - If the microparticles do not resuspend, DO NOT USE. Contact your local Abbott Representative.**
- Order tests.
- Load the ARCHITECT CA 125 II Reagent Kit on the ARCHITECT *i* System. Verify that all necessary assay reagents are present. Ensure that septa are present on all reagent bottles.

- The minimum sample volume is calculated by the system and is printed on the Orderlist report. No more than 10 replicates may be sampled from the same sample cup. To minimize the effects of evaporation, verify adequate sample cup volume is present prior to running the test.
 - Priority: 75 μ L for the first CA 125 II test plus 25 μ L for each additional test from the same sample cup.
 - \leq 3 hours on-board: 150 μ L for the first CA 125 II test plus 25 μ L for each additional CA 125 II test from the same sample cup.
 - > 3 hours on-board: additional sample volume is required. Refer to the ARCHITECT System Operations Manual, Section 5 for information on sample evaporation and volumes.
- If using primary or aliquot tubes, use the sample gauge to ensure sufficient patient specimen is present.
- ARCHITECT CA 125 II Calibrators and Controls should be mixed by gentle inversion prior to use.
 - To obtain the recommended 150 μ L volume requirements for the ARCHITECT CA 125 II Calibrators and Controls, hold the bottles vertically and dispense 4 drops of each calibrator or 4 drops of each control into each respective sample cup.
- Load samples
 - For information on loading samples, refer to the ARCHITECT System Operations Manual, Section 5.
- Press RUN. The ARCHITECT *i* System performs the following function:
 - Moves the sample to the aspiration point
 - Loads a reaction vessel (RV) into the process path
 - Aspirates and transfers sample into the RV
 - Advances the RV one position and transfers microparticles into the RV
 - Mixes, incubates and washes the reaction mixture
 - Adds conjugate to the RV
 - Mixes, incubates and washes the reaction mixture
 - Adds Pre-Trigger and Trigger Solutions
 - Measures chemiluminescent emission to determine the quantity of OC 125 defined antigen in the sample
 - Aspirates contents of RV to liquid waste and unloads RV to solid waste
 - Calculates the result
- For information on ordering patient specimens, calibrators and controls, and general operating procedures, refer to the ARCHITECT System Operations Manual, Section 5.
- It is important to follow the routine maintenance procedures defined in the ARCHITECT System Operations Manual, Section 9. If your laboratory procedures require more frequent maintenance, follow those procedures.

Specimen Dilution Procedures

- Specimens with a CA 125 II assay value exceeding 1000 U/mL are flagged with the code “ >1000.0” and may be diluted using either the Automated Dilution Protocol or the Manual Dilution Procedure.
- If using the Automated Dilution Protocol, the system performs a 1:10 dilution of the specimen and automatically calculates the concentration of the sample before dilution and reports the result.
- Manual dilutions should be performed as follows:
 - The suggested dilution for the ARCHITECT CA 125 II assay is 1:10. An additional 1:10 dilution may be made if needed.
 - For a 1:10 dilution, add 50 μ L of the patient specimen to 450 μ L of ARCHITECT *i* Multi-Assay Manual Diluent (7D82-50).
 - The operator must enter the dilution factor in the Patient or Control order screen. All assays selected for that order will be diluted. The system will use this dilution factor to automatically calculate the concentration of the sample before dilution and report the result. The dilution should be performed so that the diluted result reads greater than 20 U/mL.
- For detailed information on ordering dilutions, refer to the ARCHITECT System Operations Manual, Section 5.

Calibration

- To perform an ARCHITECT CA 125 II calibration, test calibrators A, B, C, D, E and F in duplicate. A single sample of all levels of CA 125 II controls must be tested to evaluate the assay calibration. Ensure that assay control values are within the concentration ranges specified in the control package insert. Calibrators should be priority loaded.
- Calibrator Range: 0 - 1000 U/mL.
- Once an ARCHITECT CA 125 II calibration is accepted and stored, all subsequent samples may be tested without further calibration unless:
 - A reagent kit with a new lot number is used
 - Controls are out of range
- For detailed information on how to perform an assay calibration, refer to the ARCHITECT System Operations Manual, Section 6.

QUALITY CONTROL PROCEDURES

The recommended control requirement for the ARCHITECT CA 125 II assay is a single sample of all control levels tested once every 24 hours each day of use. If the quality control procedures in your laboratory require more frequent use of controls to verify test results, follow your laboratory-specific procedures. Ensure that assay control values are within the concentration ranges specified in the package insert.

Verification of Assay Claims

For protocols to verify package insert claims, refer to the ARCHITECT System Operations Manual, Appendix B. The ARCHITECT CA 125 II assay belongs to method group 1.

RESULTS

Calculation

- The ARCHITECT CA 125 II assay utilizes a 4 Parameter Logistic Curve fit data reduction method (4PLC, Y weighted) to generate a calibration curve.

Flags

- Some results may contain information in the Flags field. For a description of the flags that may appear in this field, refer to the ARCHITECT System Operations Manual, Section 5.

LIMITATIONS OF THE PROCEDURE

- For diagnostic purposes, results should be used in conjunction with other data; e.g., symptoms, results of other tests, clinical impressions, etc.
- If the ARCHITECT CA 125 II results are inconsistent with clinical evidence, additional testing is suggested to confirm the result.
- Heterophilic antibodies in human serum can react with reagent immunoglobulins, interfering with in vitro immunoassays. Patients routinely exposed to animals or to animal serum products can be prone to this interference and anomalous values may be observed.³⁷ Additional information may be required for diagnosis.
- Specimens from patients who have received preparations of mouse monoclonal antibodies for diagnosis or therapy may contain human anti-mouse antibodies (HAMA). Such specimens may show either falsely elevated or depressed values when tested with assay kits which employ mouse monoclonal antibodies.^{38,39,40} Additional clinical or diagnostic information may be required to determine patient status.
- Patients with confirmed ovarian carcinoma may have pretreatment CA 125 assay values in the same range as healthy individuals. Elevations in circulating OC 125 defined antigen may be observed in patients with nonmalignant disease. For these reasons, a CA 125 assay value, regardless of level, should not be interpreted as absolute evidence for the presence or absence of malignant disease. The CA 125 assay value should be used in conjunction with information available from clinical evaluation and other diagnostic procedures. **The ARCHITECT CA 125 II assay should not be used as a cancer screening test.**
- Representative performance data are given in the **EXPECTED VALUES** and **SPECIFIC PERFORMANCE CHARACTERISTICS** sections. Results obtained in individual laboratories may vary.

Expected Values

The distribution of CA 125 II assay values determined in 811 specimens is shown in the table below:

Distribution of ARCHITECT CA 125 II Assay Values					
	Number of Subjects	0-35 U/mL	35.1-65 U/mL	65.1-100 U/mL	>100 U/mL
		Percent (%)			
APPARENTLY HEALTHY					
Females (Premenopausal)	99	89.9	6.1	4.0	0.0
Females (Postmenopausal)	97	99.0	1.0	0.0	0.0
MALIGNANT CONDITIONS					
Ovarian Cancer	166	49.9	14.3	4.8	32.8
Breast Cancer	50	80.0	20.0	0.0	0.0
Colorectal Cancer	50	84.0	4.0	10.0	2.0
Endometrial Cancer	25	96.0	4.0	0.0	0.0
Lung Cancer	50	60.0	18.0	10.0	12.0
NONMALIGNANT CONDITIONS					
Ovarian Disease	100	90.0	9.0	1.0	0.0
Urogenital Disease	49	83.7	14.3	2.0	0.0
Hypertension/CHD	100	88.0	11.0	0.0	1.0
Benign Endometrial	25	84.0	8.0	4.0	4.0

In this study, 94.4% of the healthy female subjects had CA 125 II assay values at or below 35.0 U/mL (mean = 16.4, SD = 13.0). It is recommended that each laboratory establish its own reference value for the population of interest.

Monitoring of Disease Status in Patients Diagnosed with Ovarian Cancer

Changes observed in serial CA 125 assay values when monitoring ovarian cancer patients should be evaluated in conjunction with other clinical methods used for monitoring ovarian cancer patients.

The effectiveness of the ARCHITECT CA 125 II assay as an aid in the monitoring of disease status in ovarian cancer patients was determined by assessing changes in CA 125 levels in serial serum samples from 63 patients compared to changes in disease status. A study involving a total of 306 observations was performed with an average number of 4.9 observations per patient. A significant change in CA 125 level was defined as at least a 10.75% increase in assay value [i.e., 2.5 times greater than the assay's total %CV (4.3%)]. Seventy-seven percent (77% or 85/111) of the positive patient samples correlated with disease progression while sixty-one percent (61% or 81/132) of serial samples showing no significant change in CA 125 assay value correlated with no progression. The total concordance in this study was sixty-eight percent (68% or 166/243). The following table presents the data in a 2 x 2 classification scheme.

Change in Disease State per Sequential Pair			
Change in CA 125 Concentration	Progression	No Progression	Total
≥10.75%	85	51	136
<10.75%	26	81	107
Total	111	132	243

The following table provides the per patient distribution. Ninety-eight percent (98% or 46/47) of the significantly increased serial samples per patient correlated with disease progression while thirty-eight percent (38% or 6/16) of serum sets showing no significant change in CA 125 level correlated with no progression. The total concordance in this study was eighty-three percent (83% or 52/63).

Change in Disease State per Patient			
Change in CA 125 Concentration	Progression	No Progression	Total
≥10.75%	46	10	56
<10.75%	1	6	7
Total	47	16	63

SPECIFIC PERFORMANCE CHARACTERISTICS

Precision

The ARCHITECT CA 125 II assay precision is ≤10% total CV. A study was performed as described per the National Committee for Clinical Laboratory Standards (NCCLS) Protocol EP5-A.⁴¹ Three defibrinated plasma-based panels were assayed, using two lots of reagents, in replicates of two at two separate times per day for 20 days on two separate instruments. Each reagent lot used a single calibration curve throughout the study. Data from this study are summarized below.*

Panel Member	Reagent Lot	Instrument	n	Mean Conc. (U/mL)	Within Run SD	%CV	Total SD	%CV
1	1	1	80	43.5	1.1	2.4	1.7	3.9
	2	2	80	49.7	0.8	1.5	0.8	1.7
2	1	1	80	303.3	9.8	3.2	11.9	3.9
	2	2	80	340.7	5.6	1.7	6.7	2.0
3	1	1	80	598.0	18.8	3.1	25.8	4.3
	2	2	80	678.3	12.4	1.8	13.5	2.0

*Representative data; results in individual laboratories may vary from these data.

Recovery

The ARCHITECT CA 125 II assay mean recovery is 100 ± 15%. A study was performed based on guidance from Tietz Textbook of Clinical Chemistry⁴² for the ARCHITECT CA 125 II assay. Known concentrations of OC 125 defined antigen were added to normal human serum samples. The concentration of CA 125 was determined using the ARCHITECT CA 125 II assay, and the resulting percent recovery was calculated. Representative data from this study are summarized in the table below*.

Sample	Endogenous Assay Value (U/mL)	OC 125 Defined Antigen Added (U/mL)	Observed CA 125 Assay Value (U/mL)	Percent Recovery**
1	36.8	165	193.7	96
		715	704.7	94
2	31.3	165	160.2	82
		715	618.6	83
3	40.2	165	186.5	91
		715	695.8	92

Average recovery across two separate spiked concentrations shown above = 90%

$$**\% \text{ Recovery} = \frac{\text{Observed CA 125 Conc. (U/mL)}}{\text{Endogenous CA 125 Conc. (U/mL)} + \text{CA 125 Added (U/mL)}} \times 100$$

*Representative data; results in individual laboratories may vary from these data.

Dilution Linearity

The ARCHITECT CA 125 II assay mean dilution linearity is 100 ± 15%. A study was performed for the ARCHITECT CA 125 II assay modeled after the National Committee for Clinical Laboratory Standards (NCCLS) Protocol EP6-P2⁴³. Samples with known elevated CA 125 concentrations were diluted with Multi-Assay Manual Diluent. The CA 125 concentration was determined for each dilution and the percent (%) recovery was calculated. Representative data from this study are summarized below*.

Sample	Final Dilution Factor	Expected Value (U/mL)	Value Obtained (U/mL)	% Recovery
1	undiluted	846.4	846.4	-
	1:1.4	604.6	631.4	104.4
	1:2	423.2	468.2	110.6
	1:3.3	256.5	282.8	110.3
	1:5	169.3	182.8	108.0
	1:10	84.6	92.7	109.5
2	undiluted	903.8	903.8	-
	1:1.4	645.6	631.6	97.8
	1:2	451.9	446.4	98.8
	1:3.3	273.9	274.0	100.1
	1:5	180.8	186.7	103.3
	1:10	90.4	95.4	105.5
3	undiluted	935.3	935.3	-
	1:1.4	668.1	645.9	96.7
	1:2	467.7	450.4	96.3
	1:3.3	283.4	284.7	100.5
	1:5	187.1	185.6	99.2
	1:10	93.5	95.8	102.4
3	undiluted	935.3	935.3	-
	1:1.4	668.1	645.9	96.7
	1:2	467.7	450.4	96.3
	1:3.3	283.4	284.7	100.5
	1:5	187.1	185.6	99.2
	1:10	93.5	95.8	102.4
3	undiluted	935.3	935.3	-
	1:1.4	668.1	645.9	96.7
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	1:5	187.1	185.6	99.2
	1:10	93.5	95.8	102.4

Average recovery across the three diluted samples above = 103.6%

$$\% \text{ Recovery} = \frac{\text{Value Obtained} \times \text{Dilution Factor}}{\text{Undiluted Concentration}} \times 100$$

*Representative data; results in individual laboratories may vary from these data.

Analytical Sensitivity

The sensitivity of the ARCHITECT CA 125 II assay is <1.0 U/mL (n=24 runs, in replicates of 10). Analytical sensitivity corresponds to the upper limit of the 95% confidence interval and represents the lowest concentration of OC 125 defined antigen that can be distinguished from zero.

Analytical Specificity

The ARCHITECT CA 125 II mean assay specificity is ≤12%. Recovery studies were performed to compare sera containing the following compounds at the indicated concentrations with control sera.*

INTERFERING SUBSTANCE

Test Compound	Test Concentration
Bilirubin	20 mg/dL
Hemoglobin	500 mg/dL
Total Protein	12 g/dL
Triglycerides	3 g/dL

CHEMOTHERAPEUTIC AGENTS

Test Compound	Test Concentration
Carboplatin	500 µg/mL
Cisplatin	165 µg/mL
Clotrimazole	0.3 µg/mL
Cyclophosphamide	500 µg/mL
Dexamethasone	10 µg/mL
Doxorubicin	1.16 µg/mL
Leucovorin	2.68 µg/mL
Melphalan	2.8 µg/mL
Methotrexate	45 µg/mL
Paclitaxel	3.5 ng/mL

*Representative data; results in individual laboratories may vary from these data.

POTENTIALLY INTERFERING CLINICAL CONDITION

The ARCHITECT CA 125 II assay was evaluated using specimens with HAMA and Rheumatoid Factor (RF) to further assess the assay specificity. Five specimens positive for HAMA and five specimens positive for RF were evaluated for % recovery with OC 125 defined antigen spiked into each specimen at 35 and 250 U/mL; mean % recovery results are summarized in the following table.*

Clinical Condition	Number of Specimens	Mean % Recovery
HAMA	10	96
RF	10	97

*Representative data; results in individual laboratories may vary from these data.

High Dose Hook

High dose hook is a phenomenon whereby very high level specimens may read within the dynamic range of the assay. For the ARCHITECT CA 125 II assay, no high dose hook effect was observed when samples containing up to approximately 180,000 U/mL of OC 125 defined antigen were assayed.

Method Comparison

The ARCHITECT CA 125 II assay method comparison correlation coefficient is ≥ 0.90 and the slope is 1.0 ± 0.15 for the full range of the assay. The ARCHITECT CA 125 II assay was compared to the Abbott AxSYM® CA 125™ assay. The results of the specimen testing are shown in the following table.*

ARCHITECT CA 125 II vs. Abbott AxSYM CA 125

Regression Method	Number of Specimens	Correlation Coefficient	Intercept (99% CI)	Slope (99% CI)
Passing-Bablok †	279**	0.985	4.0 (2.0 to 4.9)	1.06 (1.03 to 1.11)
	167***	0.967	0.4 (-0.9 to 1.8)	1.23 (1.16 to 1.30)

**Sample Range: 4.5 - 4085.9 U/mL (ARCHITECT); 2.7 - 3436.1 U/mL (AxSYM)

***Sample Range: 4.5 - 110.5 U/mL (ARCHITECT); 2.7 - 95.4 U/mL (AxSYM)

†A linear regression method with no special assumptions regarding the distribution of the samples and the measurement errors. ⁴⁴

*Representative data; results in individual laboratories may vary from these data.

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