ARCHITECT

SYSTEM

en

Total β-hCG

REF 7K78

49-3364/R3

B7K780

Read Highlighted Changes
Revised June, 2010

Total β-hCG

Customer Service: Contact your local representative or find country specific contact information on www.abbottdiagnostics.com

Package insert instructions must be carefully followed. 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	REACTION VESSELS	Reaction Vessels
IVD	*In Vitro* Diagnostic Medical Device	SAMPLE CUPS	Sample Cups
LOT	Lot Number	SEPTUM	Septum
	Expiration Date	REPLACEMENT CAPS	Replacement Caps
2°C 8°C	Store at 2-8°C	REAGENT LOT	Reagent Lot
	Consult instructions for use	SN	Serial Number
	Manufacturer		

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

Abbott

NAME

ARCHITECT Total β-hCG

INTENDED USE

The ARCHITECT Total β-hCG assay is a Chemiluminescent Microparticle Immunoassay (CMIA) for the quantitative and qualitative determination of beta human chorionic gonadotropin (β-hCG) in human serum and plasma for the early detection of pregnancy.

SUMMARY AND EXPLANATION OF TEST

Human chorionic gonadotropin (hCG) is a sialoglycoprotein with a molecular weight of approximately 46,000 daltons.[1] HCG is initially secreted by the trophoblastic cells of the placenta shortly after implantation of the fertilized ovum into the uterine wall.[2,3] The rapid rise in hCG serum levels after conception makes it an excellent marker for early confirmation and monitoring of pregnancy.

Physiologically, hCG appears to maintain the corpus luteum, thereby allowing synthesis of progesterone and estrogens that support the endometrium. As uncomplicated pregnancies progress, the placenta assumes the production of these hormones. The serum hCG levels increase to a peak concentration, then decrease and plateau. HCG circulates as the intact molecule in the serum of normal women who have an uncomplicated pregnancy. The subunits are cleaved rapidly and cleared by the kidney.[4]

The placental hormone, hCG, is similar to luteinizing hormone (LH), follicle stimulating hormone (FSH), and thyroid stimulating hormone (TSH). All are glycoproteins consisting of two noncovalently bound dissimilar subunits, designated alpha and beta, with attached carbohydrate sidechains. The alpha subunits of these glycoproteins are very similar. In contrast, the beta subunit portions determine the biological and immunochemical specificities.[5,6] The beta subunits of hCG and LH exhibit considerable homology in amino acid content. Amino acid residues specific for the beta subunit of hCG confer the immunochemical specificity.[7]

With the availability of sensitive quantitative assays for the measurement of serum β-hCG, it has been shown that hCG levels can be useful in prediction of spontaneous abortions,[8,9] aiding in the detection of ectopic pregnancy[8,10,11] and multiple gestation.[8]

BIOLOGICAL PRINCIPLES OF THE PROCEDURE

The ARCHITECT Total β-hCG assay is a two-step immunoassay to determine the presence of β-hCG 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 anti-β-hCG coated paramagnetic microparticles are combined. β-hCG present in the sample binds to the anti-β-hCG coated microparticles. After washing, anti-β-hCG 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 β-hCG 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.

REAGENTS

Reagent Kit, 100 Tests/500 Tests

NOTE: Reagent Kit configurations vary based on order.

NOTE: Some kit sizes are not available in all countries or for use on all Architect *i* Systems. Please contact your local distributor.

ARCHITECT Total β-hCG Reagent Kit (7K78)

- **MICROPARTICLES** 1 or 4 Bottle(s) (6.6 mL/27.0 mL) anti-β-hCG (mouse, monoclonal) coated Microparticles in TRIS buffer with protein (bovine) stabilizers. Preservatives: antimicrobial agents.
- **CONJUGATE** 1 or 4 Bottle(s) (2.1 mL/7.4 mL) anti-β-hCG (mouse, monoclonal) acridinium-labeled Conjugate in MES buffer with protein (bovine) stabilizers. Minimum concentration: 2.9 µg/mL. Preservative: antimicrobial agent.

Assay Diluent

ARCHITECT *i* Multi-Assay Manual Diluent (7D82-50)

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

WARNINGS AND PRECAUTIONS

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

Safety Precautions

- **CAUTION:** This product requires the handling of human specimens. It is recommended that all human sourced materials be considered potentially infectious and handled in accordance with the OSHA Standard on Bloodborne Pathogens.[12] Biosafety Level 2[13] or other appropriate biosafety practices[14,15] should be used for materials that contain or are suspected of containing infectious agents.
- 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 Total β-hCG 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.
- 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

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The ARCHITECT Total β-hCG Reagent Kit must be stored at 2-8°C 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 Total β-hCG 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 the microparticle 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 Total β-hCG Routine protocol (assay number 651) must be installed on the ARCHITECT *i*2000 from the ARCHITECT *i* Assay CD-ROM prior to performing the assay. The ARCHITECT Total β-hCG Routine protocol (assay number 651) and/or STAT protocol (assay number 030) assay files must be installed on the ARCHITECT *i* System with STAT protocol capability from the ARCHITECT *i* Assay CD-ROM prior to performing the assay. For detailed instructions on the assay file installation, refer to the ARCHITECT System Operations Manual, Section 2.
- The ARCHITECT Total β-hCG Routine protocol (two-step 18-4) is available for use on ARCHITECT *i* Systems. The ARCHITECT Total β-hCG STAT protocol (two step 4-4) is available for use on the ARCHITECT *i* System with STAT protocol capability.
- For information on printing assay parameters, refer to the ARCHITECT System Operations Manual, Section 5.
- For detailed description of system procedures, refer to the ARCHITECT System Operations Manual.
- The default result unit for the ARCHITECT Total β-hCG assay is mIU/mL. An alternative unit, IU/L, may be selected for reporting results by editing assay parameter "Result concentration units", to IU/L. The conversion factor used by the system is 1.

SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS

- Human serum (including serum collected in serum separator tubes) or plasma collected in lithium heparin, sodium heparin, or potassium EDTA may be used in the ARCHITECT Total β-hCG assay. Other anticoagulants have not been validated for use with the ARCHITECT Total β-hCG assay. Follow the tube manufacturer's processing instructions for serum or plasma collection tubes.
- The ARCHITECT *i* System does not provide the capability to verify specimen type. It is the responsibility of the operator to verify the correct specimen types are used in the ARCHITECT Total β-hCG assay.
- Inadequate centrifugation or the presence of fibrin or particulate matter in the sample may cause an erroneous result.
- Abbott Laboratories recommends the use of plasma for STAT testing. Since plasma does not require the clotting time of serum, it has a decreased likelihood of the presence of fibrin or other particulates.
- Use caution when handling patient specimens to prevent cross contamination. Use of disposable pipettes or pipette tips is recommended.
- For optimal results, 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.
- For optimal results, serum and plasma specimens should be free of fibrin, red blood cells 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, specimens should be frozen at -10°C or colder. Specimens stored frozen at -10°C or colder for 12 months showed no performance difference.
- 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.
- 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 under thermally controlled refrigerated conditions or frozen (dry ice). Prior to shipment, it is recommended that specimens be removed from the clot, serum separator or red blood cells.

PROCEDURE

Materials Provided

- 7K78 ARCHITECT Total β-hCG Reagent Kit

Materials Required but not Provided

- ARCHITECT *i* System
- ARCHITECT *i* Assay CD-ROM
- 7K78-01 ARCHITECT Total β-hCG Calibrators
- 7K78-10 ARCHITECT Total β-hCG 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**
- For information on materials required for maintenance procedures, refer to the ARCHITECT System Operations Manual, Section 9.
- Pipettes or pipette tips (optional) to deliver the volumes specified on the patient or control order screen.

Assay Procedure

- Before loading the ARCHITECT Total β-hCG 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. 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 Total β-hCG 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 cup 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 µL for the first Total β-hCG test plus 25 µL for each additional Total β-hCG test from the same sample cup
  - ≤ 3 hours onboard: 150 µL for the first Total β-hCG test plus 25 µL for each additional Total β-hCG test from the same sample cup
  - > 3 hours onboard: 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.
- To obtain the recommended volume requirements for the ARCHITECT Total β-hCG Calibrators and Controls, hold the bottles **vertically** and dispense 4 drops of each calibrator or 3 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 functions:
  - 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 detect the presence and quantity of β-hCG 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.
- For optimal performance, it is important to follow the routine maintenance procedures defined in the ARCHITECT System Operations Manual, Section 9. If your laboratory requires more frequent maintenance, follow those procedures.

Specimen Dilution Procedures

Specimens with a Total β-hCG value exceeding 15,000.00 mIU/mL are flagged with the code " >15,000.00" and may be diluted with either the Automated Dilution Protocol or the Manual Dilution Procedure.

- If using the Automated Dilution Protocol, the system performs a 1:15 dilution of the specimen and automatically calculates the concentration of the diluted specimen and reports the result.
- Manual dilutions should be performed as follows:
  - The suggested dilution for Total β-hCG is 1:15. It is recommended dilutions not exceed 1:75.
  - For a 1:15 dilution, add 20 µL of the patient specimen to 280 µL of ARCHITECT *i* Multi-Assay Manual Diluent (7D82-50).
  - The operator must enter the dilution factor in the patient or control order screen. The system will use this dilution factor to automatically calculate the concentration of the sample before dilution. This will be the reported result. The result (before dilution factor is applied) should be greater than 467.00 mIU/mL.
  - If the operator does not enter the dilution factor, the reported result will be that of the diluted sample. This result (before dilution factor is applied) should be greater than 467.00 mIU/mL.
- **NOTE:** A printed or displayed result of < 7000.00 mIU/mL (1:15 Automated Dilution Protocol) indicates the need to retest the sample at a lower dilution or undiluted. The result and interpretation should not be reported.
- For detailed information on ordering dilutions, refer to the ARCHITECT System Operations Manual, Section 5.

Calibration

- To perform an ARCHITECT Total β-hCG calibration, test Calibrators A, B, C, D, E, and F in duplicate. A single sample of all levels of Total β-hCG controls must be tested to evaluate the assay calibration. Ensure that assay control values are within the concentration ranges specified in the package insert. Calibrators should be priority loaded.
- Calibrator Range: 0.00 - 15,000.00 mIU/mL.
- Routine and STAT protocols require separate calibrations but require only one reagent kit.
- Once an ARCHITECT Total β-hCG 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 Total β-hCG 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.

Due to variation in analyte composition and/or matrices, external quality control materials and proficiency survey samples, may not elicit identical results across all hCG assays. Non-Abbott Quality Control and proficiency testing material may contain high levels of free beta-subunit hCG molecules. Non-Abbott Quality Control and proficiency testing material may generate different results when comparing a whole molecule hCG assay to a total β-hCG assay. Each laboratory needs to determine the suitability of each control material for specific immunoassays and validate the material prior to use.

Verification of Assay Claims

For protocols to verify package insert claims, refer to the ARCHITECT System Operations Manual, Appendix B. The ARCHITECT Total β-hCG assay belongs to method group 1.

RESULTS

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

Alternate Result Units

- The default result unit for the ARCHITECT Total β-hCG assay is mIU/mL. When the alternate result unit, IU/L, is selected, the conversion factor used by the system is 1.
- Conversion Formula: (Concentration in mIU/mL) x (1) = IU/L

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.

Interpretation of Results

For qualitative interpretation of the ARCHITECT Total β-hCG test results, specimens with β-hCG levels less than or equal to 5.00 mIU/mL will be reported in the INTERPRETATION field on the test results screen or printout as "NEGATIVE". Specimens with β-hCG levels greater than or equal to 25.00 mIU/mL will be reported as "POSITIVE". Specimens with β-hCG levels greater than 5.00 mIU/mL and less than 25.00 mIU/mL will be reported with concentrations only. No interpretation will be reported for these results.

LIMITATIONS OF THE PROCEDURE

- This assay is capable of detecting whole molecule (intact) hCG as well as free β-hCG subunits.
- For diagnostic purposes, hCG results should always be used in conjunction with other data; e.g., patient's medical history, symptoms, results of other tests, clinical impressions, etc. Ectopic pregnancy cannot be distinguished from normal pregnancy by hCG measurements alone.[10,11] The results from this or any other diagnostic kit should be used and interpreted only in the context of the overall clinical picture.
- If the hCG level is inconsistent with, or unsupported by, clinical evidence, results should be confirmed by an alternate hCG method. This may include the qualitative testing of urine.[29] The absence of urinary hCG may suggest a falsely elevated serum result. Results may also be confirmed by performing serial dilutions of the sample. Usually, but not always, samples that contain interfering substances exhibit nonlinear results when diluted.

- The ARCHITECT Total β-hCG assay is cleared for use in the early detection of pregnancy **only**. It is not approved for any other uses such as tumor marker screening, tumor marker monitoring, etc. and it should not be performed for any other uses.
- Infrequently, hCG levels may appear consistently elevated and could be due to, but not limited to, the presence of the following:[20,21,28,29,31,32]
  - heterophilic antibodies
  - nonspecific protein binding
  - hCG-like substances
  - trophoblastic or nontrophoblastic neoplasms
- As with any immunochemical reaction, unknown interferences from medications or endogenous substances may affect results.
- Elevated hCG levels have been associated with some abnormal physiological states (e.g., trophoblastic and nontrophoblastic neoplasms)[16,17] and the results of this test should not be used in the diagnosis of these abnormal states. There have been reports of people receiving unnecessary medical treatment and surgery, including chemotherapy and hysterectomy, when hCG results were used in the diagnosis of abnormal states.
- Interfering substances (such as heterophilic antibodies, non-specific proteins, or hCG-like substances) may falsely depress or falsely elevate results. These interfering substances may cause false results over the entire range of the assay, not just at low levels, and may indicate the presence of hCG when there is none.
- Detection of very low levels of hCG does not rule out pregnancy.[21] Low levels of hCG can occur in apparently healthy, nonpregnant subjects.[26,27] Because hCG values double approximately every 48 hours in a normal pregnancy,[21] patients with very low levels of hCG should be resampled and retested after 48 hours.
- Post menopausal specimens may elicit weak positive results due to low hCG levels unrelated to pregnancy. With a weak positive result, it is good laboratory practice to resample and retest after 48 hours, or to test with an alternate hCG method.
- Because of the high degree of sensitivity of the assay, specimens tested as positive during initial days after conception may later be negative due to natural termination of the pregnancy. Natural termination occurs in 22% of clinically unrecognized pregnancies and 31% of pregnancies overall.[30] It is good laboratory practice to resample and retest weak positive results after an additional 48 hours.
- 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 demonstrate either falsely elevated or falsely depressed results when tested with assay kits which employ Mouse Monoclonal Antibodies.[18,19] These specimens should not be tested with the Abbott ARCHITECT Total β-hCG assay.
- Heterophilic antibodies in human serum can react with reagent immunoglobulins, interfering with *in vitro* immunoassays.[20,21,32] Patients routinely exposed to animals or to animal serum products, can be prone to this interference and anomalous values may be observed. Additional information may be required for diagnosis.

EXPECTED VALUES

Because hCG is normally synthesized and secreted by cells of the placenta or its precursor, levels of the hormone in normal, nonpregnant individuals are low to undetectable.[16] Concentrations of β-hCG measured in the sera of nonpregnant individuals, as reported in the literature, are < 5 mIU/mL.[22] The concentration of β-hCG in maternal serum rises rapidly during early pregnancy. β-hCG levels between 5 mIU/mL and 25 mIU/mL may be indicative of early pregnancy.[2,23] Values for β-hCG generally peak during the first trimester and decline slowly throughout the remainder of the pregnancy.

Specimens from 200 pregnant females were categorized into four intervals based on weeks since last menstrual period (LMP; 1-10, 11-15, 16-22, 23-40) and tested using the ARCHITECT Total β-hCG assay. The results are summarized in the following table.

Weeks Post LMP (Last Menstrual Period)	n	Ranges (mIU/mL) Min	Max	2.5 percentile	97.5 percentile
1-10	50	<1.20	>225,000.00	201.64	>225,000.00
11-15	50	16,995.65	>225,000.00	22,536.49	>225,000.00
16-22	50	6,860.23	>225,000.00	8,006.62	50,238.60
23-40	50	1,583.40	65,911.38	1,599.80	49,412.65

It is recommended that each laboratory establish its own expected values.

SPECIFIC PERFORMANCE CHARACTERISTICS

Precision

The ARCHITECT Total β-hCG assay is designed to have a precision of < 10% (total CV). A study based on guidance from Clinical and Laboratory Standards Institute (CLSI, formerly NCCLS) document EP5-T2[24] was performed for the ARCHITECT Total β-hCG assay. A three member processed human serum based panel was assayed, using two lots of reagents, in replicates of two at two separate times per day for 20 days. Data from this study are summarized in the following table.*

Panel Member	Reagent Lot	Instrument	n	Mean Conc. Value (mIU/mL)	Within Run SD	Within Run %CV	Total SD	Total %CV
1	1	1	80	21.07	0.984	4.7	1.027	4.9
1	1	2	80	24.97	0.880	3.5	1.115	4.5
1	2	1	80	25.22	1.027	4.1	1.134	4.5
1	2	2	80	24.75	0.819	3.3	1.096	4.4
2	1	1	80	735.93	14.917	2.0	18.144	2.5
2	1	2	80	772.66	12.161	1.6	14.671	1.9
2	2	1	80	750.99	13.812	1.8	16.065	2.1
2	2	2	80	754.87	11.762	1.6	12.592	1.7
3	1	1	80	5,063.95	84.801	1.7	107.073	2.1
3	1	2	80	5,116.40	72.066	1.4	86.052	1.7
3	2	1	80	4,976.44	98.439	2.0	109.379	2.2
3	2	2	80	5,052.13	59.656	1.2	79.602	1.6

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

Recovery

The ARCHITECT Total β-hCG assay is designed to have a mean recovery of 100 ± 10% when analyzing specimens spiked with known amounts of β-hCG. A known concentration of hCG (1000.45 mIU/mL) was added to eight aliquots of human serum. The concentration of β-hCG was determined using the ARCHITECT Total β-hCG assay and the resulting percent recovery was calculated. The percent recovery of the ARCHITECT Total β-hCG assay ranged from 104.1% to 108.8% with an average of 106.7%. This is representative data; results in individual laboratories may vary from these data.

Dilution Verification

The accuracy of the ARCHITECT Total β-hCG Automated Dilution Protocol was evaluated using human serum specimens. Results from specimens tested in replicates of two using the 1:15 Automated Dilution Protocol were compared to results from the same specimens diluted 1:15 with the ARCHITECT *i* Multi-Assay Manual Diluent. Performance was evaluated as percent recovery and was calculated using the following equation:*

$$\% \text{ Recovery} = \frac{\text{Automated Dilution β-hCG Conc. (mIU/mL)}}{\text{Manual Dilution β-hCG Conc. (mIU/mL)}} \times 100$$

Specimen	Manual Dilution β-hCG Conc. (mIU/mL)	Automated Dilution β-hCG Conc. (mIU/mL)	% Recovery
A	98,488.05	102,152.34	103.7
B	27,534.10	27,199.45	98.8
C	47,730.65	46,994.66	98.5
D	13,413.25	13,438.39	100.2
E	10,391.55	9,463.34	91.1

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

Analytical Sensitivity

The ARCHITECT Total β-hCG assay is designed to have an analytical sensitivity of ≤ 1.2 mIU/mL. Analytical Sensitivity is defined as the concentration at two standard deviations from the mean RLU value of the ARCHITECT Total β-hCG Calibrator A (0.00 mIU/mL) and represents the lowest measurable concentration of β-hCG that can be distinguished from zero.

Specificity

The ARCHITECT Total β-hCG assay is designed to have a mean analytical specificity of < 10% cross reactivity with FSH, LH, and TSH. Aliquots of human serum containing β-hCG were supplemented with 150 mIU/mL FSH, 250 mIU/mL LH, and 100 µIU/mL TSH and assayed for β-hCG. The cross reactivity was calculated as a percent interference and was shown to be less than 10% for each cross reactant.

Carryover

Carryover from a sample containing 1,000,000 mIU/mL β-hCG to an adjacent 0 mIU/mL β-hCG sample was less than 7.5 mIU/mL β-hCG.

NOTE: Please be aware that individual samples may exhibit elevated concentration due to build up of proteins on the sample pipettor probe. For further troubleshooting information, refer to the ARCHITECT System Operations Manual, Section 10.

Interference

The ARCHITECT Total β-hCG assay is designed to have a potential interference from hemoglobin, bilirubin, triglycerides and protein of < 10% at the levels indicated below.

- Hemoglobin - < 10% at 500 mg/dL
- Bilirubin - < 10% at 20 mg/dL
- Triglycerides - < 10% at 3000 mg/dL
- Protein - < 10% at 2 g/dL and 12 g/dL

Accuracy by Correlation

The ARCHITECT Total β-hCG assay is designed to have a slope of 1.0 ± 0.15 and a correlation coefficient (r) of > 0.95 within the range of 0 - 15,000 mIU/mL when compared to the AxSYM Total β-hCG assay.

A study was performed where specimens were tested using the ARCHITECT Total β-hCG assay and the AxSYM Total β-hCG assay. Data from this study were analyzed using least squares and Passing-Bablok [25] regression methods and are summarized in the following table.*

Abbott ARCHITECT Total β-hCG vs. Abbott AxSYM Total β-hCG

Method	Number of Specimens	AxSYM Range (mIU/mL)	Inter-cept	Slope	Correlation Coefficient
Least Squares	93	2 - 1,000[1]	3.40	1.00	0.945
Linear	192	2 - 15,000[2]	58.34	0.89	0.980
Regression	471	2 - 200,000[3]	-672.30	0.82	0.986
Passing-	93	2 - 1,000[1]	0.48	1.08	0.945
Bablock Linear	192	2 - 15,000[2]	1.88	0.94	0.980
Regression**	471	2 - 200,000[3]	19.84	0.80	0.986

In this evaluation, serum specimens tested on the ARCHITECT Total β-hCG assay ranged from: [1]1.99 to 1262.17 mIU/mL, [2]1.99 to 14934.93 mIU/mL, [3]1.99 to 180620.00 mIU/mL.

* Representative data; variables such as differences in sampling size and sample population may impact the correlation of the assay; therefore, results in individual laboratories may vary from these data.

** A linear regression method with no special assumptions regarding the distribution of the samples and the measurement errors.[25]

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