Access
Immunoassay Systems

BECKMAN COULTER

TOTAL βhCG (5th IS)

Standardized to the WHO 5th International Standard for Chorionic Gonadotropin

REF A85264

FOR USE WITH TEST NAMES: HCG5, HCG5d and d-CG5

Intended Use

The Access Total βhCG (5th IS) assay is a paramagnetic particle, chemiluminescent immunoassay for the quantitative determination of total βhCG levels in human serum and plasma using the Access Immunoassay Systems. This assay is intended for use as an aid in the early detection of pregnancy.

Summary and Explanation

Human chorionic gonadotropin (hCG) is a glycoprotein hormone, produced by the placenta, with structural similarity to the pituitary hormones FSH, TSH, and LH. The alpha subunits (MW 15,000–20,000 daltons) are common to all of these hormones but the beta subunits differ, and confer immunological and biological specificity. βhCG (MW 25,000–30,000 daltons) shares several peptide sequences with βLH, but has a unique carboxyl terminal region.[1]

Shortly after implantation of a fertilized ovum into the uterine wall, the trophoblast begins to produce hCG, which maintains steroid secretions of the corpus luteum until the placenta can do so.[2] hCG can be detected after implantation; concentrations double approximately every 1.5 to 3 days for the first six weeks and then continue to rise until the end of the first trimester, gradually falling to a lower level for the remainder of the pregnancy.[3,4] After delivery, hCG returns to < 5 mIU/mL (IU/L) and is usually undetectable several days postpartum.

The hormone is an excellent marker for pregnancy. Healthy, non-pregnant individuals have low (< 5 mIU/mL [IU/L]) to undetectable hCG levels; however, hCG, originating from the pituitary gland, can be found at detectable levels in peri- and post-menopausal women.[5] During pregnancy, unusually low or rapidly declining levels may indicate an abnormal condition such as an ectopic pregnancy or impending spontaneous abortion.[6]

Principles of the Procedure

The Access Total βhCG (5th IS) assay is a sequential two-step immunoenzymatic ("sandwich") assay. A sample is added to a reaction vessel along with a citrate buffer. After an initial incubation, rabbit anti-βhCG alkaline phosphatase conjugate and paramagnetic particles coated with goat anti-mouse IgG: mouse monoclonal anti-βhCG complexes are added. The hCG binds to the immobilized monoclonal anti-βhCG on the solid phase while, at the same time, the rabbit anti-βhCG alkaline phosphatase conjugate reacts with different antigenic sites on the hCG. After incubation in a reaction vessel, materials bound to the solid phase are held in a magnetic field while unbound materials are washed away. Then, the chemiluminescent substrate Lumi-Phos* 530 is added to the vessel and light generated by the reaction is measured with a luminometer. The light production is directly proportional to the concentration of total βhCG in the sample. The amount of analyte in the sample is determined from a stored, multi-point calibration curve.

Product Information

Access Total βhCG (5th IS) Reagent Pack

Cat. No. A85264: 100 determinations, 2 packs, 50 tests/pack

- Provided ready to use.
- Store upright and refrigerate at 2 to 10°C.
- Refrigerate at 2 to 10°C for a minimum of two hours before use on the instrument.
- Stable until the expiration date stated on the label when stored at 2 to 10°C.

- Stable at 2 to 10°C for 28 days after initial use.
- Signs of possible deterioration are a broken elastomeric layer on the pack or control values out of range.
- If the reagent pack is damaged (e.g., broken elastomer), discard the pack.
- All antisera are polyclonal unless otherwise indicated.
- A fine suspension of particulate may be observed in the R1b well of the reagent pack. The presence of this particulate has demonstrated no effect on assay performance.

R1a:	Paramagnetic particles coated with goat anti-mouse IgG: mouse monoclonal anti-βhCG complexes suspended in TRIS buffered saline, with surfactant, bovine serum albumin (BSA), < 0.1% sodium azide, and 0.1% ProClin** 300.
R1b:	Protein (goat, murine, and recombinant) diluted in citrate buffered saline, with surfactant, < 0.1% sodium azide, and 0.1% ProClin 300.
R1c:	Rabbit anti-βhCG alkaline phosphatase (recombinant) conjugate diluted in MES buffered saline, with surfactant, BSA, protein (rabbit), < 0.1% sodium azide, and 0.25% ProClin 300.

Warnings and Precautions

- For *in vitro* diagnostic use.
- Patient samples and blood-derived products may be routinely processed with minimum risk using the procedure described. However, handle these products as potentially infectious according to universal precautions and good clinical laboratory practices, regardless of their origin, treatment, or prior certification. Use an appropriate disinfectant for decontamination. Store and dispose of these materials and their containers in accordance with local regulations and guidelines.
- Human source material used in the preparation of the reagent has been tested and found negative or non-reactive for Hepatitis B, Hepatitis C (HCV), and Human Immunodeficiency Virus (HIV-1 and HIV-2). Because no known test method can offer complete assurance that infectious agents are absent, handle reagents and patient samples as if capable of transmitting infectious disease.[7]
- Sodium azide may react with lead and copper plumbing to form highly explosive metal azides. On disposal of liquids, flush with a large volume of water to prevent azide build-up.[8]
- Xi. Irritant: 0.25% ProClin 300.

R 43: May cause sensitization by skin contact.

S 28-37: After contact with skin, wash immediately with plenty of soap and water. Wear suitable gloves.

- The Material Safety Data Sheet (MSDS) is available upon request.

Specimen Collection and Preparation

1. Serum and plasma (lithium heparin) are the recommended samples. Urine is not a recommended sample type.
2. The role of preanalytical factors in laboratory testing has been described in a variety of published literature.[9,10,11] To minimize the effect of preanalytical factors observe the following recommendations for handling, processing, and storing blood samples:[10]
   - Collect all blood samples observing routine precautions for venipuncture.
   - Allow serum samples to clot completely before centrifugation in an upright position. Clotting may be slowed at cooler temperatures or if the patient is on anticoagulant therapy.
   - Keep tubes stoppered at all times.
   - Physically separate serum or plasma from contact with cells as soon as possible.
   - Store samples tightly stoppered at room temperature (15 to 30°C) for no longer than eight hours.

- If the assay will not be completed within eight hours, refrigerate the samples at 2 to 8°C.
- If the assay will not be completed within 48 hours, or for shipment of samples, freeze at -20°C or colder.
- Frozen specimens can be stored up to six months before testing.[12]
- Thaw samples only once.

3. Use the following guidelines when preparing specimens:
   - Ensure residual fibrin and cellular matter have been removed prior to analysis.
   - Follow blood collection tube manufacturer's recommendations for centrifugation.
4. Each laboratory should determine the acceptability of its own blood collection tubes and serum separation products. Variations in these products may exist between manufacturers and, at times, from lot-to-lot.

Materials Provided

R1 Access Total βhCG (5th IS) Reagent Packs

Materials Required But Not Provided

1. Access Total βhCG (5th IS) Calibrators
   Provided at zero and approximately 6, 35, 195, 620 and 1350 mIU/mL (IU/L).
   Cat. No. B11754
2. Quality Control (QC) materials: commercial control material.
3. Access Substrate
   Cat. No. 81906
4. **Access 2, UniCel DxC 600i:**
   Access Wash Buffer II, Cat. No. A16792
   UniCel DxI 600, UniCel DxI 800, UniCel DxC 880i, UniCel DxC 860i, UniCel DxC 680i, UniCel DxC 660i:
   UniCel DxI Wash Buffer II, Cat. No. A16793
   UniCel DxI Access Immunoassay Systems Wash Buffer II, Cat. No. A79784 (Diluent pack for use with the UniCel DxI system onboard dilution feature.)

Procedural Comments

1. Refer to the appropriate system manuals and/or Help system for a specific description of installation, start-up, principles of operation, system performance characteristics, operating instructions, calibration procedures, operational limitations and precautions, hazards, maintenance, and troubleshooting.
2. Mix contents of new (unpunctured) reagent packs by gently inverting pack several times before loading on the instrument. Do not invert open (punctured) packs.
3. Use twenty-five (25) µL of sample for each determination in addition to the sample container and system dead volumes when requesting the **HCG5** assay. Use twenty (20) µL of sample in addition to the sample container and system dead volumes for each determination run with the DxI system onboard dilution feature (test name: **d-CG5**). Use six (6) µL of sample for each determination in addition to the sample container and system dead volumes when requesting the **HCG5d** assay. Refer to the appropriate system manuals and/or Help system for the minimum sample volume required.
4. The system default unit of measure for sample results is mIU/mL. To change sample reporting units to the International System of Units (SI units), IU/L, refer to the appropriate system manuals and/or Help system. To manually convert concentrations to the International System, multiply mIU/mL by multiplication factor 1.

Procedure

Refer to the appropriate system manuals and/or Help system for information on managing samples, configuring tests, requesting tests, and reviewing test results.

- Select **HCG5** as the test name for assaying samples containing total βhCG concentrations up to the concentration of the Access Total βhCG (5th IS) S5 calibrator.

- UniCel DxI users may use either the UniCel DxI onboard dilution feature (Test name **d-CG5**) or the special dilution feature (Test name **HCG5d**) for assaying samples containing total βhCG concentrations greater than the Access Total βhCG (5^{th} IS) S5 calibrator.
- The same reagent pack is used for all assays.

Calibration Details

An active calibration curve is required for all tests. For the Access Total βhCG (5^{th} IS) assay, calibration is required every 28 days. Refer to the appropriate system manuals and/or Help system for information on calibration theory, configuring calibrators, calibrator test request entry, and reviewing calibration data.

Quality Control

Quality control materials simulate the characteristics of patient samples and are essential for monitoring the system performance of immunochemical assays. Because samples can be processed at any time in a "random access" format rather than a "batch" format, quality control materials should be included in each 24-hour time period.[13] Include commercially available quality control materials that cover at least two levels of analyte. More frequent use of controls or the use of additional controls is left to the discretion of the user based on good laboratory practices or laboratory accreditation requirements and applicable laws. Follow manufacturer's instructions for reconstitution and storage. Each laboratory should establish mean values and acceptable ranges to assure proper performance. Quality control results that do not fall within acceptable ranges may indicate invalid test results. Examine all test results generated since obtaining the last acceptable quality control test point for this analyte. Refer to the appropriate system manuals and/or Help system for information about reviewing quality control results.

Results

Patient test results are determined automatically by the system software. The amount of analyte in the sample is determined from the measured light production by means of the stored calibration data. Patient test results can be reviewed using the appropriate screen. Refer to the appropriate system manuals and/or Help system for complete instructions on reviewing sample results.

Limitations of the Procedure

1. This assay is only intended for use as an aid in the early detection of pregnancy.
2. Samples can be accurately measured within the analytical range of the lower limit of quantitation and the highest calibrator value (0.6 to approximately 1350 mIU/mL [IU/L]).
   - If a sample contains less than the lower limit of quantitation for the assay, report the results as less than that value (i.e., < 0.6 mIU/mL [IU/L]).
   - If a sample contains more than the stated value of the highest Access Total βhCG (5^{th} IS) Calibrator (S5), report the result as greater than that value (i.e., > 1350 mIU/mL [IU/L]). Alternatively, use one of the dilution options below.

Onboard Dilution Feature for use on UniCel DxI systems:

Samples containing hCG concentrations greater than the concentration of the Access Total βhCG (5^{th} IS) S5 calibrator can be processed using the DxI onboard dilution feature. The DxI system onboard dilution feature automates the dilution process, using one volume of sample with 199 volumes of Wash Buffer II from the UniCel DxI Access Immunoassay Systems Diluent Pack (Cat. No. A79784) allowing samples to be quantitated up to approximately 270,000 mIU/mL.

Test Name	Reportable Range (mIU/mL)	Sample Volume Required
d-CG5	1050 to ~270,000	20 µL

Note: The system reports the results adjusted for the dilution. If the sample contains less than 1050 mIU/mL, the system will report results as < 1050 mIU/mL. Any neat sample reading < 1050 mIU/mL in the **d-CG5** assay should be retested in the **HCG5** assay.

Dil-Total βhCG (5th IS) Special Dilution Feature for use on Access 2 and UniCel DxI systems:

Samples containing hCG concentrations greater than the concentration of the Access Total βhCG (5th IS) S5 calibrator can be processed using the Special Dilution Feature. When **HCG5d** is requested, the system automatically dilutes the sample using Wash Buffer II and reads the resulting dose off the **HCG5** calibration curve.

Test Name	Reportable Range (mIU/mL)	Sample Volume Required
HCG5d	1050 to ~270, 000	6 µL

Note: The system multiplies by the dilution factor defined in the software to calculate final test results. If the sample contains less than 1050 mIU/mL, the system will report results as < 1050 mIU/mL. Any neat sample resulted as < 1050 mIU/mL in the **HCG5d** assay should be retested in the **HCG5** assay.

Manual off-line pre-dilution

- Dilute one volume of sample with 199 volumes of Wash Buffer II (1/200).
- Type in the pre-dilution factor when entering the test request. Order the **HCG5** test.
- The system will automatically multiply the result by the pre-dilution factor and report that value.
- If the pre-dilution option was not entered when requesting the test, multiply the calculated value by the dilution factor 200 after assaying the diluted sample using the **HCG5** assay.
- Refer to the appropriate system manuals and/or Help system for additional instructions on processing pre-diluted samples.

3. DO NOT reuse small sample volumes that have been resident on the analyzer for more than one hour.
4. For assays employing antibodies, the possibility exists for interference by heterophile antibodies in the patient sample. Patients who have been regularly exposed to animals or have received immunotherapy or diagnostic procedures utilizing immunoglobulins or immunoglobulin fragments may produce antibodies, e.g. HAMA, that interfere with immunoassays. Additionally, other heterophile antibodies such as human anti-goat antibodies may be present in patient samples.[14,15] Such interfering antibodies may cause erroneous results. Carefully evaluate the results of patients suspected of having these antibodies.
5. Other potential interferences in the patient sample could be present and may cause erroneous results in immunoassays. Some examples that have been documented in literature include rheumatoid factor, endogenous alkaline phosphatase, fibrin, and proteins capable of binding to alkaline phosphatase.[16] Carefully evaluate the results of patients suspected of having these types of interferences.
6. The role of preanalytical factors in laboratory testing has been described in a variety of published literature.[9,10,11] Following blood collection tube manufacturers' specimen collection and handling recommendations will help to reduce preanalytical error.
7. Low levels of hCG β-core fragment do not react with the Access Total βhCG (5th IS) assay. Interference from high levels of hCG β-core fragment has not been tested.
8. Automatic dilutions of serum samples (by Dil-Total βhCG (5th IS) or onboard dilution) have the potential of generating individual results with bias > 15%. For representative data see Dilution Recovery section.
9. The Access Total βhCG (5th IS) results should be interpreted in light of the total clinical presentation of the patient, including: symptoms, clinical history, data from additional tests, and other appropriate information.
10. The Access Total βhCG (5th IS) assay has no discernible "hook" effect up to 1,000,000 mIU/mL.

11. If the total βhCG result is not consistent with clinical presentation, results should be confirmed by an alternate hCG method or a urine-based assay.[17]
12. Trophoblastic or nontrophoblastic neoplastic conditions and post-menopausal status should be considered before interpreting results.

Expected Values

1. Each laboratory should establish its own reference ranges to assure proper representation of specific populations.
2. Total βhCG concentrations were measured in human serum samples collected from apparently healthy non-pregnant females using the Access Total βhCG (5th IS) assay. Concentrations of total βhCG measured in 100% of samples were determined to be ≤ 11.6 mIU/mL (IU/L). The observed ranges and 95th percentile of total βhCG concentrations are shown in the table below.

Reference Population (Non-Pregnant Females)	N	Median (mIU/mL)	Range (mIU/mL)	95th Percentile (mIU/mL), [95% CI]
≥ 18 and < 40 years	132	0	0 – 0.6	0.3 [0.2 – 0.4]
≥ 40 years	141	0	0 – 3.1	1.5 [1.1 – 2.9]
Post-menopausal***	134	2.8	0.1 – 11.6	7.7 [6.4 – 10.4]

***Post-menopausal status confirmed using circulating FSH and estradiol levels.

Healthy, non-pregnant individuals typically have low (< 5 mIU/mL [IU/L]) to undetectable hCG levels; however, hCG can rise to detectable levels in peri- and post-menopausal women.[5] hCG results between 5 mIU/mL and 25 mIU/mL may be indicative of early pregnancy but should be interpreted in light of the total clinical presentation of the patient.[18]

Specific Performance Characteristics

Methods Comparison

A comparison of 224 serum values using the Access Total βhCG (5th IS) assay (Cat. No. A85264) on the UniCel DxI 800 Immunoassay System and a commercially available immunoassay system yielded the following statistical data using Passing-Bablok regression:

n	Range of Observations † (mIU/mL)	Intercept (mIU/mL)	Slope (95% CI)	Correlation Coefficient (r)
224	3.2 – 1095.9	2.87	1.04 (1.02 – 1.06)	0.99

†Observed concentration range of the Access Total βhCG (5th IS) assay.

Linearity

Based on CLSI EP6-A,[19] one high sample (> 1350 mIU/mL) and one low sample (< 0.2 mIU/mL) were mixed to make 13 evenly distributed sample concentrations. Four replicates of the 13 mixed samples, 8 replicates of the low sample and 8 replicates of the high sample were run on a single DxI 800 system. The Access Total βhCG (5th IS) assay was designed to be linear, with a maximum deviation from linearity of ≤ 10.0% for samples > 3.9 mIU/mL, and ≤ 0.39 mIU/mL (1.0 SD) for samples ≤ 3.9 mIU/mL. One study, analyzed using a polynomial regression method demonstrated a maximum deviation from linearity of 7.5% for samples > 3.9 mIU/mL, and 0.09 mIU/mL for samples ≤ 3.9 mIU/mL.

Dilution Recovery

Five serum samples and five lithium heparin plasma samples slightly above the Access Total βhCG (5th IS) S5 calibrator (approximately 1350 mIU/mL) were diluted 1/200 with Wash Buffer II. When run on the UniCel DxI 800 Immunoassay System, the average percent recoveries of the individual plasma samples ranged from 99-109% and the average percent recoveries of the individual serum samples ranged from 95-114%.

Representative data using five individual serum samples with eight replicates for each sample at approximately 1,500 mIU/mL is shown below:

Serum Sample	Dil-Total βhCG (5^{th} IS) Assay Average Recovery (%)	Dil-Total βhCG (5^{th} IS) Assay Recovery Range (%)	Onboard Dilution Assay Average Recovery (%)	Onboard Dilution Assay Recovery Range (%)
Sample 1	109	99 – 118	114	106 – 123
Sample 2	109	96 – 119	112	106 – 119
Sample 3	97	88 – 108	100	92 – 108
Sample 4	95	84 – 102	100	93 – 107
Sample 5	96	84 – 103	102	93 – 108

Imprecision

The Access Total βhCG (5^{th} IS) assay exhibits total imprecision ≤ 10.0% CV at concentrations greater than 3.9 mIU/mL, and total Standard Deviation (SD) ≤ 0.39 mIU/mL at concentrations ≤ 3.9 mIU/mL. Two separate studies, one using human serum samples and one using human plasma samples, involved a total of 40 runs, with two replicates per run, over 20 days. The following data were calculated based on CLSI EP5-A2[20] guidelines.

Serum Sample	Grand Mean (n=80) (mIU/mL)	Within Run		Between Run		Total Imprecision	
		SD (mIU/mL)	%CV	SD (mIU/mL)	%CV	SD (mIU/mL)	%CV
Sample 1	0.6	0.07	N/A	0.11	N/A	0.13	N/A
Sample 2	4.1	0.11	2.7	0.24	6.0	0.27	6.6
Sample 3	24.0	0.68	2.8	0.30	1.2	0.74	3.1
Sample 4	106.7	1.97	1.8	2.60	2.4	3.26	3.1
Sample 5	791.3	24.24	3.1	40.81	5.2	47.46	6.0
Sample 6	1116.1	38.19	3.4	53.29	4.8	65.56	5.9
Sample 7	208,778	7,482	3.6	9,400	4.5	12,014	5.8

Plasma Sample	Grand Mean (n=80) (mIU/mL)	Within Run		Between Run		Total Imprecision	
		SD (mIU/mL)	%CV	SD (mIU/mL)	%CV	SD (mIU/mL)	%CV
Sample 1	1.4	0.06	N/A	0.06	N/A	0.08	N/A
Sample 2	4.7	0.23	4.8	0.20	4.2	0.30	6.4
Sample 3	675.0	19.81	2.9	21.45	3.2	29.20	4.3
Sample 4	1112.3	43.35	3.9	58.09	5.2	72.48	6.5

Analytical Specificity/Interferences

Samples containing interferents at the concentrations listed below do not affect the concentration of total βhCG reported.

Substance	Concentration Added
Acetaminophen	20 mg/dL
Acetylsalicylic Acid	65 mg/dL
Bilirubin (conjugated and unconjugated)	40 mg/dL
Hemoglobin	500 mg/dL
Heparin (Low Molecular Weight)	7200 U/dL
Total Protein (Human Serum Albumin)	6 g/dL
Ibuprofen	50 mg/dL
Triglycerides (Intralipid)	3 g/dL
Multi-vitamin	0.9% (v/v)

The following table describes the cross-reactivity of the Access Total βhCG (5th IS) assay when substances that are similar in structure to hCG were added to a patient sample with an approximate hCG concentration of 2.9 mIU/mL. This study was performed on a UniCel DxI 800 Immunoassay System and values were calculated as described in CLSI EP7-A2.[21] When the concentrations of cross-reactants below were added to the Access Total βhCG (5th IS) Calibrator S0 (zero) the result was below the limit of detection.

Substance	Concentration Added (mIU/mL)	hCG Concentration Without Cross-Reactant (mIU/mL)	hCG Concentration With Cross-Reactant (mIU/mL)
hLH	103	2.25	2.10
hFSH	1000	2.30	2.23
hTSH	1	2.44	2.53
hCG α-subunit	500	2.95	2.87

The Access Total βhCG (5th IS) assay recognizes intact hCG, the β subunit of hCG, nicked intact hCG and nicked βhCG isoforms. The free α subunit and β-core fragment yield no detectable response.

Limit of Blank

The Access Total βhCG (5th IS) assay is designed to have a Limit of Blank (LoB) of ≤ 0.5 mIU/mL (IU/L). In one study, LoB was tested using a protocol based on CLSI EP17-A2.[22] A total of 156 replicates of a zero analyte sample, the Access Total βhCG (5th IS) Calibrator S0, were measured in 12 runs using multiple reagent packs and calibrator lots on multiple UniCel DxI 800 Immunoassay Systems. This study determined the LoB for Access Total βhCG (5th IS) to be 0.1 mIU/mL (IU/L).

Limit of Detection

The Access Total βhCG (5th IS) assay is designed to have a Limit of Detection (LoD) of ≤ 0.5 mIU/mL (IU/L). In one study, LoD was tested using a protocol based on CLSI EP17-A2.[22] Thirty-six replicates from 6 low-level samples were measured using multiple reagent packs and calibrator lots in 12 runs on multiple UniCel DxI 800 Immunoassay Systems. This study determined the LoD for Access Total βhCG (5th IS) to be 0.2 mIU/mL (IU/L).

Limit of Quantitation

The Access Total βhCG (5th IS) assay is designed to have a Limit of Quantitation (LoQ) of ≤ 0.6 mIU/mL (IU/L). In one study, LoQ was tested using a protocol based on CLSI EP17-A2.[22] Ten replicates of eight low-level samples were measured using three reagent pack lots and one calibrator lot in six runs on multiple UniCel DxI 800 Immunoassay Systems. LoQ was determined as the lowest concentration which met the design requirements of 20% CV and recovery of +/- 0.1 mIU/mL for three reagent lots when compared to the WHO 5th International Standard for Chorionic Gonadotropin (NIBSC Code 07/364). This study determined the LoQ for Access Total BhCG (5th IS) to be 0.6 mIU/mL (IU/L).

Access
Immunoassay Systems

BECKMAN COULTER

TOTAL βhCG (5th IS) CALIBRATORS

REF B11754

Intended Use

The Access Total βhCG (5th IS) Calibrators are intended to calibrate the Access Total βhCG (5th IS) assay for the quantitative determination of total βhCG levels in human serum and plasma using the Access Immunoassay Systems.

Summary and Explanation

Quantitative assay calibration is the process by which samples with known analyte concentrations (i.e., assay calibrators) are tested to measure the response. The mathematical relationship between the measured responses and the known analyte concentrations establishes the calibration curve. This mathematical relationship, or calibration curve, is used to convert Relative Light Unit (RLU) measurements of samples of unknown concentration to specific quantitative analyte concentrations.

Traceability

The measurand (analyte) in the Access Total βhCG (5th IS) Calibrators is traceable to the WHO 5th International Standard for Chorionic Gonadotropin (NIBSC Code 07/364).[23,24] Traceability process is based on EN ISO 17511.
The assigned values were established using representative samples from this lot of calibrator and are specific to the assay methodologies of the Access reagents. Values assigned by other methodologies may be different. Such differences, if present, may be caused by inter-method bias.

Product Information

Access Total βhCG (5th IS) Calibrators
Cat. No. B11754: S0–S5, 4.0 mL/vial

- Provided ready to use.
- Store upright and freeze at -20°C or colder.
- Stable until the expiration date stated on the label when stored at -20°C or colder.
- Thaw at room temperature. Mix contents by gently inverting before use. Avoid bubble formation.
- Thaw calibrators no more than 15 times.
- Return calibrators to -20°C or colder after each use.
- Vial is stable at -20°C or colder for 120 days after initial use.
- Signs of possible deterioration are control values out of range.
- Refer to calibration card for exact concentrations.

S0:	Buffered bovine serum albumin (BSA) matrix with surfactant, < 0.1% sodium azide, and 0.5% ProClin** 300. Contains 0.0 mIU/mL (IU/L) hCG.
S1, S2, S3, S4, S5:	hCG at levels of approximately 6, 35, 195, 620 and 1350 mIU/mL (IU/L), respectively, in buffered BSA matrix with surfactant, < 0.1% sodium azide, and 0.5% ProClin 300.
Calibration Card:	1

Warnings and Precautions

- For *in vitro* diagnostic use.

- Patient samples and blood-derived products may be routinely processed with minimum risk using the procedure described. However, handle these products as potentially infectious according to universal precautions and good clinical laboratory practices, regardless of their origin, treatment, or prior certification. Use an appropriate disinfectant for decontamination. Store and dispose of these materials and their containers in accordance with local regulations and guidelines.
- Human source material used in the preparation of the reagent has been tested and found negative or non-reactive for Hepatitis B, Hepatitis C (HCV), and Human Immunodeficiency Virus (HIV-1 and HIV-2). Because no known test method can offer complete assurance that infectious agents are absent, handle reagents and patient samples as if capable of transmitting infectious disease.[7]
- Human source material was purified from the urine of pregnant women and should be treated as potentially infectious.[7]
- Sodium azide may react with lead and copper plumbing to form highly explosive metal azides. On disposal of liquids, flush with a large volume of water to prevent azide build-up.[8]
- Xi. Irritant: 0.5% ProClin 300.

R 43: May cause sensitization by skin contact.
S 28-37: After contact with skin, wash immediately with plenty of soap and water. Wear suitable gloves.

- The Material Safety Data Sheet (MSDS) is available upon request.

Procedure

Refer to the appropriate system manuals and/or Help system for information on calibration theory, configuring calibrators, calibrator test request entry, and reviewing calibration data.

Calibration Details

The Access Total βhCG (5th IS) Calibrators are provided at 6 levels – zero and approximately 6, 35, 195, 620 and 1350 mIU/mL. Assay calibration is required every 28 days.

Calibrators run in duplicate.

Limitations of the Procedure

If there is evidence of microbial contamination or excessive turbidity in a reagent, discard the vial.

References

1 Cole LA and Butler SA, "Structure, Synthesis, Secretion, and Function of hCG." Human Chorionic Gonadotropin (HCG), Burlington, MA: Elsevier, Inc., 2010; p 49-53.

2 Kardana A, et al. The heterogeneity of hCG. Endocrinology 1991; 129: 1541-1567.

3 Sturgeon CM and McAllister EJ. Analysis of hCG: clinical applications and assay requirements. Ann Clin Biochem 1998; 35: 460-491.

4 Vaitukaitis JL. Recent progress in hormone research 1976; 32: 289.

5 Gronowski AM, et al. Use of Serum FSH to Identify Perimenopausal Women with Pituitary hCG. Clin Chem 2008; 54 (4): 652-656.

6 Sokolove PJ, Faix JD. Agreement of intact and beta chain-specific HCG assays in abnormal pregnancy. Journal of Clinical Immunoassay, Fall, 1991; 14, No. 3: 196-199.

7 HHS Publication, 5th ed., December 2009. Biosafety in Microbiological and Biomedical Laboratories. Available http://www.cdc.gov/biosafety/publications/bmbl5/index.htm

8 DHHS (NIOSH) Publication No. 78-127, August 1976. Current Intelligence Bulletin 13 - Explosive Azide Hazard. Available http://www.cdc.gov/niosh.

9 Blood Human Chorionic Gonadotropin (hCG) Assays: What Laboratorians Should Know About False-Positive Results. Food and Drug Administration Center for Devices and Radiological Health Medical Device Reporting, 2009. Available http://www.fda.gov/MedicalDevices/Safety/AlertsandNotices/TipsandArticlesonDeviceSafety/ucm109390.htm. Accessed July 2013.

10 Approved Guideline – Procedures for the Handling and Processing of Blood Specimens for Common Laboratory Tests, H18-A4. 2010. Clinical and Laboratory Standards Institute.

11 Approved Standard – Sixth Edition, Procedures for the Collection of Diagnostic Blood Specimens by Venipuncture, H3-A6. 2007. Clinical and Laboratory Standards Institute.

12 O'Connor JF, et al. Recent advances in the chemistry and immunochemistry of human chorionic gonadotropin: impact on clinical measurements, Endocrine Reviews 1994; 15, No. 5: 650-683.

13 Cembrowski GS, Carey RN. Laboratory quality management: QC⇌QA. ASCP Press, Chicago, IL, 1989.

14 Kricka L. Interferences in immunoassays – still a threat. Clin Chem 2000; 46: 1037–1038.

15 Bjerner J, et al. Immunometric assay interference: incidence and prevention. Clin Chem 2002; 48: 613–621.

16 Lingwood D, Ballantyne JS. Alkaline phosphatase-immunoglobulin conjugate binds to lipids in vitro, independent of antibody selectivity. Journal of Immunological Methods 2006; 311: 174–177.

17 Cole, LA. Phantom hCG and Phantom Choriocarcinoma. Gynecol Oncol, 1998; 71: 325–9.

18 Tietz NW, Textbook of Clinical Chemistry and Molecular Diagnostics, 4th ed. 2006. p. 364, 2261.

19 Approved Guideline – Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach, EP6-A. April 2003. Clinical and Laboratory Standards Institute.

20 Approved Guideline – Evaluation of Precision Performance of Quantitative Measurement Methods, EP5-A2. August 2004. Clinical and Laboratory Standards Institute.

21 Approved Guideline – Interference Testing in Clinical Chemistry, EP7-A2. November 2005. Clinical and Laboratory Standards Institute.

22 Approved Guideline – Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures, EP17-A2. June 2012. Clinical and Laboratory Standards Institute.

23 5th WHO IS Chorionic Gonadotropin (NIBSC Code 07/364) Instructions for Use. Available http://www.nibsc.ac.uk/products/biological_reference_materials/product_catalogue/detail_page.aspx?catid=07/364

24 Burns et al, WHO international collaborative study of the proposed 5th [fifth] international standard for chorionic gonadotrophin. Geneva: World Health Organization, 2009. Available http://apps.who.int/iris/handle/10665/70154

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