ARCHITECT

SYSTEM

en

i Carbamazepine

REF 1P36

G2-6210/R02

B1P360

Read Highlighted Changes
Revised August 2012

i Carbamazepine

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	CONTROL NO.	Control Number
IVD	*In Vitro* Diagnostic Medical Device	REACTION VESSELS	Reaction Vessels
LOT	Lot Number	REAGENT LOT	Reagent Lot
SN	Serial number	REPLACEMENT CAPS	Replacement Caps
	Expiration Date	SEPTUMS	Septums
2°C 8°C	Store at 2-8°C	SAMPLE CUPS	Sample Cups
	Consult instructions for use.	WARNING: SENSITIZER	WARNING: May cause an allergic reaction.
	Manufacturer	GTIN	Global Trade Item Number
		PRODUCT OF GERMANY	Product of Germany

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

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NAME

ARCHITECT *i*Carbamazepine

INTENDED USE

The ARCHITECT *i*Carbamazepine assay is an *in vitro* chemiluminescent microparticle immunoassay (CMIA) for the quantitative measurement of carbamazepine, an anticonvulsant drug, in human serum or plasma (collected in lithium heparin, sodium heparin, dipotassium EDTA or sodium EDTA tubes) on the ARCHITECT *i* System with *STAT* protocol capability. The measurements obtained are used in monitoring levels of carbamazepine to help ensure appropriate therapy.

SUMMARY AND EXPLANATION OF TEST

Carbamazepine is an iminostilbine derivative structurally related to the tricyclic antidepressants.[1,2] Carbamazepine is used in the treatment of both generalized tonic-clonic and simple and complex partial seizures because of its inhibition of repetitive firing of neurons.[2]

About 75% of the carbamazepine in plasma is protein bound.[2,3] Carbamazepine is metabolized by hepatic oxidases, primarily CYP3A4, yielding the 10,11-epoxide.[2] This metabolite is also pharmacologically active and is found in plasma and tissues at concentrations up to 50% of the parent drug.[2] The 10,11-epoxide is further metabolized to inactive compounds that are excreted in the urine, mostly as glucuronides.[1,2]

BIOLOGICAL PRINCIPLES OF THE PROCEDURE

The ARCHITECT *i*Carbamazepine assay is a one-step immunoassay for the quantitative measurement of carbamazepine in human serum or plasma using CMIA technology, with flexible assay protocols, referred to as Chemiflex.

In the ARCHITECT *i*Carbamazepine assay, the sample, anti-carbamazepine coated paramagnetic microparticles, and carbamazepine acridinium-labeled conjugate are combined to create a reaction mixture. The anti-carbamazepine coated microparticles bind to carbamazepine present in the sample and to the carbamazepine acridinium-labeled conjugate. After washing, pre-trigger and trigger solutions are added to the reaction mixture. The resulting chemiluminescent reaction is measured as relative light units (RLUs). An indirect relationship exists between the amount of carbamazepine in the sample and the RLUs detected by the ARCHITECT *i* System optics.

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

REAGENTS

Reagent Kit, 100 Tests

ARCHITECT *i*Carbamazepine Reagent Kit (1P36)

- **MICROPARTICLES** 1 Bottle (6.6 mL) Anti-carbamazepine (mouse, monoclonal) coated microparticles in MES buffer with protein (bovine) stabilizer. Minimum concentration: 0.04% solids. Preservative: ProClin 300.
- **CONJUGATE** 1 Bottle (10.0 mL) Carbamazepine acridinium-labeled conjugate in MES buffer. Minimum concentration: 4 ng/mL. Preservative: ProClin 300.

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 0.35 N sodium hydroxide.

ARCHITECT *i* Wash Buffer

- **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 be handled in accordance with the OSHA Standard on Bloodborne Pathogens.[4] Biosafety Level 2[5] or other appropriate biosafety practices[6,7] should be used for materials that contain or are suspected of containing infectious agents.
- The following warnings and precautions apply to these components:
  - Microparticles
  - Conjugate

WARNING:	Contains methylisothiazolones
H317	May cause an allergic skin reaction.
Prevention	
P261	Avoid breathing mist/vapours/spray.
P272	Contaminated work clothing should not be allowed out of the workplace.
P280	Wear protective gloves/protective clothing/ eye protection.
Response	
P302+P352	IF ON SKIN: Wash with plenty of water.
P333+P313	If skin irritation or rash occurs: Get medical advice/attention.
P363	Wash contaminated clothing before reuse.
	This material and its container must be disposed of in a safe way.

- Safety Data Sheets are available at www.abbottdiagnostics.com or contact your local representative.
- For a detailed discussion of safety precautions during system operation, refer to the ARCHITECT System Operations Manual, Section 8.

Handling Precautions

- Do not use reagents kits beyond the expiration date.
- Do not freeze reagents.
- **Do not pool reagents within a kit or between reagent kits.**
- Before loading the ARCHITECT *i*Carbamazepine Reagent Kit on the system for the first time, the microparticle bottle requires mixing to resuspend microparticles that may have settled during shipment. For microparticle mixing instructions, refer to the **PROCEDURE, Assay Procedure** section of this package insert.
- **Septums MUST be used to prevent reagent evaporation and contamination and to ensure reagent integrity. Reliability of assay results cannot be guaranteed if septums 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 the 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, and 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 *i*Carbamazepine 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, the reagents are stable until the expiration date.
- The ARCHITECT *i*Carbamazepine Reagent Kit may be stored on board the ARCHITECT *i* System with *STAT* protocol capability for a maximum of 30 days. After 30 days, the reagent kit must be discarded. Recalibration may be required to obtain maximum onboard reagent stability. For information on tracking onboard 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 septums 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.** For information on unloading reagents, refer to the ARCHITECT System Operations Manual, Section 5.

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 are invalid and samples must be retested. Assay recalibration may be necessary. For troubleshooting information, refer to the ARCHITECT System Operations Manual, Section 10.

INSTRUMENT PROCEDURE

- The ARCHITECT *i*Carbamazepine assay is designed for use on the ARCHITECT *i* System with *STAT* protocol capability.
- The ARCHITECT *i*Carbamazepine assay file must be installed on the ARCHITECT *i* System with *STAT* protocol capability before performing the assay. For detailed information on assay file installation and 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.
- The default result unit for the ARCHITECT *i*Carbamazepine assay is µg/mL.
  - When the alternate result unit µmol/L is selected, the conversion factor used by the system is 4.23.
    - Conversion Formula:
      (concentration in µg/mL) x (4.23) = µmol/L
  - When the alternate result unit mg/L is selected, the conversion factor used by the system is 1.00.
    - Conversion Formula:
      (concentration in µg/mL) x (1.00) = mg/L

SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS

Specimen Types

The specimen collection tubes listed below were verified for use with the ARCHITECT *i*Carbamazepine assay. Other specimen collection tubes have not been tested with this assay.

Glass	Plastic
• Serum • Sodium EDTA	• Serum • Lithium heparin • Sodium heparin • Dipotassium EDTA

- 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 types are used in the ARCHITECT *i*Carbamazepine assay.

Specimen Conditions

- Do not use specimens with the following conditions:
  - heat-inactivated
  - pooled
  - grossly hemolyzed
  - obvious microbial contamination
- Performance has not been established for the use of cadaveric specimens or body fluids other than human serum or plasma.
- **For accurate results, serum and plasma specimens should be free of fibrin, red blood cells, and other particulate matter.** Serum specimens from patients receiving anticoagulant or thrombolytic therapy may contain fibrin due to incomplete clot formation.
- Use caution when handling patient specimens to prevent cross contamination. Use of disposable pipettes or pipette tips is recommended.
- For optimal results, inspect all specimens for bubbles. Remove bubbles with an applicator stick before analysis. Use a new applicator stick for each specimen to prevent cross contamination.

Preparation for Analysis

- Follow the tube manufacturer's processing instructions for serum and plasma collection tubes. Gravity separation is not sufficient for specimen preparation.
- Mix thawed specimens thoroughly by low-speed vortexing or by inverting 10 times. Visually inspect the specimens. If layering or stratification is observed, continue mixing until specimens are visibly homogeneous.
- To ensure consistency in results, specimens must be transferred to a centrifuge tube and centrifuged at >10,000 RCF (Relative Centrifugal Force) for 10 minutes before testing if
  - they contain fibrin, red blood cells, or other particulate matter or
  - they were frozen and thawed.
- Centrifuged specimens with a lipid layer on the top must be transferred to a sample cup or secondary tube. Care must be taken to transfer only the clarified specimen without the lipemic material.
- Transfer clarified specimen to a sample cup or secondary tube for testing.

Storage

- Specimens may be stored on or off the clot or red blood cells for
  - up to 24 hours at room temperature or
  - up to 7 days at 2-8°C.
- If testing will be delayed more than 7 days, remove serum or plasma from the clot or red blood cells and store at -20°C or colder.
- Avoid more than 3 freeze/thaw cycles.

Shipping

- Before shipping specimens, it is recommended that specimens be removed from the clot or red blood cells.
- When shipping specimens, package and label specimens in compliance with applicable state, federal, and international regulations covering the transport of clinical specimens and infectious substances.
- Specimens may be shipped ambient, at 2-8°C (wet ice), or frozen (dry ice). Do not exceed the storage time limitations listed above.

PROCEDURE

Materials Provided

- 1P36 ARCHITECT *i*Carbamazepine Reagent Kit

Materials Required but not Provided

- ARCHITECT *i* System with *STAT* protocol capability
- Architect *i*Carbamazepine Assay File, may be obtained from:
  - Architect *i* System e-Assay CD-ROM found on www.abbottdiagnostics.com
  - Architect *i* System Assay CD-ROM
- 1P36-01 ARCHITECT *i*Carbamazepine Calibrators
- Commercially available control material containing carbamazepine
- 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 specified volumes.

For information on materials required for maintenance procedures, refer to the ARCHITECT System Operations Manual, Section 9.

Assay Procedure

- Before loading the ARCHITECT *i*Carbamazepine Reagent Kit on the system for the first time, the microparticle bottle requires mixing to resuspend microparticles that may have settled during shipment. After the first time the microparticles have been loaded, no further mixing is required.
  - **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.
  - **If the microparticles do not resuspend, DO NOT USE. Contact your Abbott representative.**
  - Once the microparticles have been resuspended, place a septum on the bottle. For instructions about placing septums on bottles, refer to the **Handling Precautions** section of this package insert.
- Load the ARCHITECT *i*Carbamazepine Reagent Kit on the ARCHITECT *i* System with *STAT* protocol capability
  - Verify that all necessary reagents are present.
  - Ensure that septums are present on all reagent bottles.
- Order calibration, if necessary.
  - For information on ordering calibrations, refer to the ARCHITECT System Operations Manual, Section 6.
- Order tests.
  - For information on ordering patient specimens and controls and for general operating procedures, refer to the ARCHITECT System Operations Manual, Section 5.
  - For additional information, refer to the **RESULTS, Flags** section of this package insert.
- 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 before running the test.
  - Priority: 80 µL for the first *i*Carbamazepine test plus 30 µL for each additional *i*Carbamazepine test from the same sample cup.
  - ≤ 3 hours on-board: 150 µL for the first *i*Carbamazepine test plus 30 µL for each additional *i*Carbamazepine test from the same sample cup.
  - > 3 hours on-board: replace with a fresh sample (patient specimens, controls, and calibrators).
  - If using primary or aliquot tubes, use the sample gauge to ensure sufficient patient specimen is present.
- Prepare calibrators and controls.
  - Mix the ARCHITECT *i*Carbamazepine Calibrators by gentle inversion before use.
  - To obtain the recommended volume requirements for the ARCHITECT *i*Carbamazepine Calibrators, hold the bottles **vertically**, and dispense a minimum of 5 drops of each calibrator into each respective sample cup.
  - Follow the manufacturer's instructions for preparation of commercially available control material.
- Load samples
  - For information on loading samples, refer to the ARCHITECT System Operations Manual, Section 5.
- Press RUN.
- For additional information on principles of operation, refer to the ARCHITECT System Operations Manual, Section 3.
- For optimal performance, it is important to perform routine maintenance as described in the ARCHITECT System Operations Manual, Section 9. Perform maintenance more frequently when required by laboratory procedures.

Specimen Dilution Procedures

- Specimens with a carbamazepine concentration greater than 20.00 µg/mL will be flagged as > 20.00 and may be diluted using the Manual Dilution Procedure below.

Manual Dilution Procedure

- The suggested dilution for the ARCHITECT *i*Carbamazepine assay is 1:4.
  - For example, add 50 µL of the patient specimen to 150 µL of ARCHITECT *i*Carbamazepine Calibrator A.
- The result should be greater than 2.00 µg/mL before the dilution factor is applied.
- 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.
- For detailed information on ordering dilutions, refer to the ARCHITECT System Operations Manual, Section 5.

Calibration

- To perform an ARCHITECT *i*Carbamazepine calibration, test Calibrators A, B, C, D, E, and F in duplicate. The calibrators should be priority loaded.
- Calibration Range: 0.00 to 20.00 µg/mL.
- To evaluate the calibration of this assay using commercially available controls, a single sample of all levels of controls should be tested.
  - Order controls as described in the **Assay Procedure** section.
  - Ensure that assay control values are within the established ranges.
- Once an ARCHITECT *i*Carbamazepine 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 *i*Carbamazepine assay is that a single sample of each control be tested once every 24 hours each day of use. If quality control procedures in your laboratory require more frequent use of controls to verify test results, follow your laboratory-specific procedures or your federal, state, and/or local accrediting agency requirements or regulations.
- Commercial controls should be used according to the guidelines and recommendations of the control manufacturer. In addition, each laboratory should establish its own concentration ranges for new control lots at each control level employed. These ranges should be established according to your laboratory quality control policy and/or any local, state, and/or federal regulations or accreditation requirements. Concentration ranges provided in the control package insert should be used only for guidance.
- For any control material in use, the laboratory should ensure that the matrix of the control material is suitable for use in the assay per the assay package insert.
- If a control is out of its established range, the associated test results may be invalid and should be retested per laboratory procedures. Recalibration may be indicated.
- Refer to published guidelines for information or general control recommendation, for example Clinical and Laboratory Standards Institute (CLSI) Document C24-A3[8] or other published guidelines, for general quality control recommendations.

Verification of Assay Claims

For protocols to verify package insert claims, refer to the ARCHITECT System Operations Manual, Appendix B. The ARCHITECT *i*Carbamazepine assay belongs to method group 2.

RESULTS

The ARCHITECT *i*Carbamazepine assay uses a 4 Parameter Logistic Curve fit (4PLC, Y-weighted) data reduction method 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.

Measuring Interval (Reportable Range)

The measuring interval of the ARCHITECT *i*Carbamazepine assay is 2.00 µg/mL to 20.00 µg/mL. Refer to the **SPECIFIC PERFORMANCE CHARACTERISTICS, Measuring Interval** section of this package insert. When using the manual dilution procedure, the assay can report values up to 80.0 µg/mL.

LIMITATIONS OF THE PROCEDURE

- If the carbamazepine results are inconsistent with clinical evidence, additional testing is suggested to confirm the result.
- For diagnostic purposes, results should be used in conjunction with other data; *e.g.*, symptoms, results of other tests, clinical impressions, etc.
- Specimens from patients who have received preparations of mouse monoclonal antibodies for diagnosis or therapy may contain human anti-mouse antibodies (HAMA).[9,10] Specimens containing HAMA may produce anomalous values when tested with assay kits such as ARCHITECT *i*Carbamazepine that employ mouse monoclonal antibodies.[9]
- Heterophilic antibodies in human serum can react with reagent immunoglobulins, interfering with *in vitro* immunoassays.[11] Patients routinely exposed to animals or to animal serum products can be prone to this interference and anomalous results may be observed. Additional information may be required for diagnosis.
- Refer to the **SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS** section of this package insert for specimen limitations.

EXPECTED VALUES

Plasma concentrations between 4 and 12 µg/mL of carbamazepine have been associated with optimal seizure control in adults.[12] Toxicity associated with carbamazepine therapy is generally relatively minor. The most serious problem is concerned with carbamazepine's ability to suppress bone marrow function. This serious toxic effect, potentially leading to aplastic anemia, is rare.[13] More frequently encountered side effects such as drowsiness, incoordination, vertigo, and diplopia are dose-related and not of a life threatening nature.[14,15]

Refer to the drug manufacturer's package insert or the Physicians' Desk Reference (PDR) for proper drug dosage and for carbamazepine measurement sampling times.

SPECIFIC PERFORMANCE CHARACTERISTICS

Data in the **SPECIFIC PERFORMANCE CHARACTERISTICS** section were generated using the ARCHITECT *i* 2000_{SR} System.

Assay results obtained in individual laboratories may vary from data presented.

Precision

The ARCHITECT *i*Carbamazepine assay is designed to have an imprecision of ≤ 7% Total CV for samples with carbamazepine concentrations across the range of 2.00 µg/mL to 20.00 µg/mL.

Within-Laboratory Precision

A study was performed based on guidance from the National Committee for Clinical Laboratory Standards (NCCLS) document EP5-A2.[16] Testing was conducted at Abbott Laboratories using two lots of ARCHITECT *i*Carbamazepine Reagent Kits, two lots of ARCHITECT *i*Carbamazepine Calibrators, one lot of commercially available controls (Bio-Rad Liquichek Therapeutic Drug Monitoring Controls), and three instruments. Three levels of controls and four levels of human serum panels were assayed in a minimum of two replicates at two separate times per day for 20 different days. Each reagent lot used a single calibration curve throughout the study. The data are summarized in the following table.

Sample	Instrument	Reagent Lot	n	Mean (µg/mL)	Within Run SD	Within Run %CV	Total SD	Total %CV
Control Level 1	1	1	120	3.94	0.074	1.9	0.112	2.9
		2	120	3.91	0.074	1.9	0.125	3.2
	2	1	120	4.01	0.060	1.5	0.095	2.4
		2	120	4.05	0.077	1.9	0.103	2.6
	3	1	120	3.85	0.055	1.4	0.059	1.5
		2	120	3.85	0.049	1.3	0.053	1.4
Control Level 2	1	1	120	9.61	0.242	2.5	0.318	3.3
		2	120	9.55	0.198	2.1	0.302	3.2
	2	1	120	9.75	0.184	1.9	0.225	2.3
		2	120	9.99	0.168	1.7	0.220	2.2
	3	1	120	9.43	0.109	1.2	0.113	1.2
		2	120	9.56	0.140	1.5	0.146	1.5
Control Level 3	1	1	120	15.12	0.263	1.7	0.503	3.3
		2	120	15.18	0.381	2.5	0.582	3.8
	2	1	120	15.36	0.422	2.7	0.501	3.3
		2	120	15.60	0.271	1.7	0.443	2.8
	3	1	120	14.71	0.204	1.4	0.244	1.7
		2	120	14.96	0.194	1.3	0.241	1.6
Panel 1	1	1	120	1.89	0.042	2.2	0.073	3.9
		2	120	1.84	0.043	2.3	0.066	3.6
	2	1	120	1.91	0.040	2.1	0.060	3.2
		2	120	1.87	0.049	2.6	0.056	3.0
	3	1	120	1.80	0.026	1.5	0.030	1.7
		2	120	1.78	0.031	1.7	0.034	1.9
Panel 2	1	1	120	2.23	0.055	2.5	0.087	3.9
		2	120	2.20	0.046	2.1	0.076	3.4
	2	1	120	2.28	0.050	2.2	0.060	2.7
		2	120	2.24	0.055	2.4	0.068	3.0
	3	1	120	2.14	0.030	1.4	0.034	1.6
		2	120	2.11	0.036	1.7	0.038	1.8
Panel 3	1	1	120	12.52	0.260	2.1	0.457	3.6
		2	120	12.48	0.238	1.9	0.472	3.8
	2	1	120	12.75	0.298	2.3	0.385	3.0
		2	120	12.96	0.280	2.2	0.357	2.8
	3	1	120	12.20	0.194	1.6	0.230	1.9
		2	120	12.37	0.192	1.6	0.260	2.1
Panel 4	1	1	120	18.65	0.390	2.1	0.767	4.1
		2	120	18.84	0.480	2.5	0.800	4.2
	2	1	120	18.76	0.366	1.9	0.429	2.3
		2	120	19.01	0.373	2.0	0.410	2.2
	3	1	120	17.97	0.291	1.6	0.309	1.7
		2	120	18.30	0.221	1.2	0.259	1.4

Recovery

The ARCHITECT *i*Carbamazepine assay is designed to have a mean percent recovery of 90% to 110% for samples with carbamazepine concentrations ranging from 4 µg/mL to 12 µg/mL.

A study was performed with 12 specimens obtained from patients receiving carbamazepine therapy. The specimens were spiked with additional carbamazepine to create test samples with carbamazepine concentrations within the range of 6 µg/mL to 18 µg/mL. The samples were tested using the ARCHITECT *i*Carbamazepine assay on one instrument, and the resulting percent recovery was calculated. The individual percent recovery ranged from 94.1% to 110.0%, and the mean percent recovery was 101.4%.

Sensitivity

Limit of Quantitation

The ARCHITECT *i*Carbamazepine assay is designed to have a Limit of Quantitation (LoQ) of ≤ 2.0 µg/mL.

Based on guidance from the NCCLS document EP17-A,[17] a study was performed with five zero-level samples and five samples with carbamazepine concentrations of approximately 0.3, 0.5, 0.7, 1.0, and 1.5 µg/mL. These samples were tested in five separate runs over a minimum of 3 days using two reagent lots and two instruments. The LoQ for the ARCHITECT *i*Carbamazepine assay was 0.30 µg/mL.

Limit of Blank and Limit of Detection

In the same study, the Limit of Blank (LoB) and Limit of Detection (LoD) were determined. The LoB was 0.06 µg/mL and the LoD was 0.13 µg/mL.

Linear Range

Based on guidance from the NCCLS document EP6-A,[18] a study was performed to establish the linear range of the ARCHITECT *i*Carbamazepine assay. Three dilution series were prepared as follows: a high carbamazepine sample (> 20 to ≤ 26 µg/mL) was combined in specific ratios with a low carbamazepine sample (< 2 µg/mL). The three dilution series, including the low-level and high-level samples, were tested using the ARCHITECT *i*Carbamazepine assay.

Using an absolute deviation from linearity of ≤ 10% for samples with concentrations between 2 µg/mL and 20 µg/mL and of ≤ 0.20 µg/mL for samples with carbamazepine concentrations less than 2 µg/mL, a linear range of 0.56 µg/mL to 22.34 µg/mL was established for the ARCHITECT *i*Carbamazepine assay.

Measuring Interval

Measuring Interval is defined as the range of values in µg/mL which meets the limits of acceptable performance for both imprecision and bias for an undiluted sample. For the verification studies described in this package insert, the range was 2.00 µg/mL to 20.00 µg/mL.

Interference

Potentially Interfering Endogenous Substances

Based on guidance from the CLSI document EP7-A2,[19] potentially interfering endogenous substances were evaluated to determine whether carbamazepine concentrations were affected when using the ARCHITECT *i*Carbamazepine assay. The endogenous substances listed below were spiked into samples with different levels of carbamazepine (approximately 4 µg/mL and 12 µg/mL). The samples were assayed, and the carbamazepine concentrations of the spiked samples were compared to control samples. The data are summarized in the following table.

Potentially Interfering Endogenous Substance	Minimum Interferent Concentration	% Interference [a]	
		4 µg/mL	12 µg/mL
Bilirubin	20 mg/dL	3.9%	2.9%
Conjugated Bilirubin	20 mg/dL	4.7%	5.2%
Hemoglobin	500 mg/dL	6.2%	4.1%
Total Protein	12 g/dL	3.0%	9.1%
Triglycerides	3000 mg/dL	-2.0%	-0.1%

[a] $$\% \text{ Interference} = \frac{\text{Test Result - Control Result}}{\text{Control Result}} \times 100$$

Potentially Interfering Clinical Conditions

Potentially interfering clinical conditions were evaluated to determine whether carbamazepine concentrations were affected when using the ARCHITECT *i*Carbamazepine assay. Specimens from individuals with the clinical conditions listed below were divided into three samples. Two of the samples were spiked to different levels of carbamazepine (approximately 4 µg/mL and 12 µg/mL). All samples were assayed, and the results of the samples spiked with carbamazepine were compared to samples that were not spiked with carbamazepine. The data are summarized in the following table.

Potentially Interfering Clinical Condition	n	% Interference Range [a] (Individual)	
		4 µg/mL	12 µg/mL
Human Anti-Mouse Antibodies (HAMA)	12	-6.6% to 7.4%	-7.8% to 13.9%
Rheumatoid Factor	12	-4.5% to 8.7%	-6.7% to 8.5%
Heterophilic Antibody	12	-2.5% to 9.8%	-8.4% to 4.3%

[a] $$\% \text{ Interference} = \frac{\text{(Spiked Result − Unspiked Result) − Added Carbamazepine Concentration}}{\text{Added Carbamazepine Concentration}} \times 100$$

Potentially Interfering Drugs

Based on guidance from the CLSI document EP7-A2,[19] potentially interfering drugs were evaluated to determine whether carbamazepine concentrations were affected when using the ARCHITECT *i*Carbamazepine assay. Potentially interfering drugs were spiked into normal human serum at two levels of carbamazepine (approximately 4 µg/mL and 12 µg/mL) and into normal human serum (0 µg/mL). The samples were assayed, and the results of the spiked samples with carbamazepine concentrations of 4 µg/mL and 12 µg/mL were compared to the spiked normal human serum. The data are summarized in the following table.

Test Compound	Test Compound Conc. (µg/mL)	Carbamazepine Concentration (µg/mL)						
		0	4			12		
		Conc. Diff.[a]	Conc. Diff.[a]	% Interf.[b]	% Cross-React.[c]	Conc. Diff.[a]	% Interf.[b]	% Cross-React.[c]
5-(p-Hydroxy-phenyl)-5-phenylhydantoin	1000	0.00	-0.10	-2.3	-0.0	-0.11	-0.9	-0.0
10-Hydroxy-carbamazepine	22	0.14	0.26	6.2	1.2	-0.05	-0.4	-0.2
Acetaminophen	200	-0.01	0.01	0.2	0.0	0.16	1.4	0.1
Acetylcysteine	150	0.02	0.26	6.1	0.2	0.61	5.1	0.4
Acetylsalicylic Acid	1000	0.00	0.07	1.8	0.0	0.03	0.2	0.0
Amitriptyline	100	0.00	0.01	0.4	0.0	-0.05	-0.4	-0.0
Amobarbital	50	0.00	-0.19	-4.4	-0.4	-0.59	-4.8	-1.2
Ampicillin-Na	100	-0.03	-0.22	-5.2	-0.2	-0.43	-3.7	-0.4
Ascorbic Acid	30	0.03	0.22	5.1	0.7	0.74	6.3	2.5
Carbamazepine-10,11-epoxide	6	0.21	0.24	5.5	3.9	0.30	2.6	5.1
Cefoxitin	2500	0.05	0.30	7.1	0.0	0.90	7.6	0.0
Cetirizine dihydrochloride	3	-0.01	-0.02	-0.5	-0.7	0.12	1.0	3.9
Chlordiazepoxide	30	0.00	0.03	0.6	0.1	0.17	1.4	0.6
Chlorpromazine	100	0.00	0.01	0.2	0.0	0.01	0.1	0.0
Clonazepam	12	0.02	0.07	1.8	0.6	-0.43	-3.5	-3.6
Cyclosporine	5	0.01	0.23	5.3	4.6	0.10	0.8	2.0
Desipramine	5	0.00	0.00	0.1	0.1	0.02	0.1	0.3
Diazepam	25	0.00	0.00	0.0	0.0	-0.16	-1.3	-0.6
Eslicarbazepine	170	0.00	0.01	0.3	0.0	-0.01	-0.1	-0.0
Ethosuximide	1000	0.02	-0.02	-0.5	-0.0	-0.07	-0.5	-0.0
Ethotoin	50	0.00	-0.01	-0.3	-0.0	0.14	1.2	0.3
Glutethimide	50	0.00	-0.02	-0.4	-0.0	0.00	0.0	0.0
Hydroxyzine dihydrochloride	1	0.06	0.08	1.9	8.2	-0.15	-1.2	-14.7
Ibuprofen	500	0.02	0.10	2.5	0.0	0.18	1.5	0.0
Imipramine	200	0.02	0.00	0.0	0.0	0.00	0.0	0.0
K-Dobesilate (hydroquinone-sulfonic acid potassium salt)	200	-0.01	-0.13	-3.0	-0.1	-0.05	-0.5	-0.0
Levodopa	20	0.00	0.29	7.0	1.4	0.87	7.6	4.4
Mephenytoin	150	0.01	-0.04	-1.0	-0.0	-0.02	-0.2	-0.0
Methsuximide	50	0.00	0.05	1.3	0.1	-0.07	-0.6	-0.1
Methyldopa sesquihydrate	20	0.01	0.18	4.3	0.9	0.42	3.5	2.1
Metronidazole	200	-0.01	-0.03	-0.8	-0.0	0.05	0.4	0.0
Nortriptyline	50	0.04	0.08	2.0	0.2	0.21	1.8	0.4
Oxcarbazepine	10	0.14	0.20	4.8	2.0	0.13	1.1	1.3
p-Hydroxy-phenobarbital	50	-0.00	0.00	0.0	0.0	-0.05	-0.4	-0.1
Phenobarbital	50	-0.01	-0.06	-1.4	-0.1	0.28	2.4	0.6
Phenothiazine	200	0.00	-0.06	-1.6	-0.0	-0.06	-0.5	-0.0
Phenylbutazone	400	0.00	-0.14	-3.4	0.0	-0.54	-4.4	-0.1
Phenytoin	1000	0.02	0.09	2.1	0.0	0.29	2.5	0.0
Primidone	1000	0.01	0.05	1.2	0.0	-0.05	-0.4	-0.0
Probenecid	500	0.00	-0.04	-0.9	-0.0	-0.20	-1.7	-0.0
Promethazine	1000	0.03	-0.35	-8.0	-0.0	-0.35	-3.0	-0.0
Rifampicin	60	0.00	-0.13	-3.1	-0.2	0.19	1.6	0.3
Secobarbital	50	-0.01	0.05	1.3	0.1	-0.04	-0.3	-0.1
Tetracycline	50	-0.00	-0.02	-0.6	-0.0	0.06	0.5	0.1
Theophylline	100	0.00	-0.07	-1.5	-0.1	0.08	0.6	0.1
Valproic acid	1000	0.01	0.21	5.0	0.0	0.56	4.8	0.1

[a] Diff (Interference) = mean/median test concentration − mean/median reference concentration

[b] % Interf = [Diff / (mean/median reference concentration)] *100

[c] % Cross-reactivity = (Diff / cross-reactant concentration) *100

Method Comparison

The ARCHITECT *i*Carbamazepine assay is designed to have a slope of 1.0 ± 0.10 and a correlation coefficient (r) of ≥ 0.90 for samples across the range of 2.00 µg/mL to 20.00 µg/mL carbamazepine when compared to AxSYM Carbamazepine. A correlation study was performed based on guidance from the NCCLS Document EP9-A2[20] using the Passing-Bablok regression method to compare the ARCHITECT *i*Carbamazepine assay to the AxSYM Carbamazepine assay using serum specimens (n = 128). The data are summarized in the following table.

Concentration Range (µg/mL)		Correlation Coefficient (r)		Inter-cept (µg/mL)	95% CI[b]	Slope	95% CI[b]
ARCHITECT	AxSYM	r	95% CL[a]				
2.22 - 18.93	2.73 - 14.75	0.934	0.907	-0.68	(-1.19, -0.18)	1.06	(0.99, 1.14)

[a] = 95% CL = Confidence Limit (Lower, One-sided)
[b] = CI = Confidence Interval

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The following U.S. Patents are relevant to the ARCHITECT System or its components. There are other such patents and patent applications in the United States and worldwide.

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