ARCHITECT iDigoxin

REF 1P32-27

en

iDigoxin
1P32
G10802R02
B1P3X0

Revised January 2018.

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.

WARNING: Digoxin values for specimens from patients who have received DIGIBIND or DIGIFAB therapy may be impacted. See **SPECIFIC PERFORMANCE CHARACTERISTICS, Interfering Substances** section.

NAME

ARCHITECT iDigoxin

INTENDED USE

The ARCHITECT iDigoxin assay is an *in vitro* chemiluminescent microparticle immunoassay (CMIA) for the quantitative measurement of digoxin in human serum or plasma on the ARCHITECT iSystem with STAT protocol capability. The measurements obtained are used to aid in the diagnosis and treatment of digoxin overdose and in monitoring levels of digoxin to help ensure appropriate therapy.

SUMMARY AND EXPLANATION OF THE TEST

Digoxin is a potent cardiac glycoside prescribed for the treatment of patients suffering from congestive heart failure[1] or from some types of cardiac arrhythmias.[2] Monitoring of serum or plasma digoxin levels is performed because the drug has a low therapeutic ratio (a small difference between therapeutic and tissue toxic levels) and because the symptoms of drug overdose may resemble the original condition for which the drug was prescribed.[3] Also, digoxin dosage may require adjustment when renal function is impaired[3] or when drugs known to alter the pharmacokinetics of digoxin (e.g., quinidine, verapamil, or amiodarone) are coadministered.[4] Monitoring serum or plasma digoxin levels along with other clinical data can aid the physician in adjusting patient dosage to achieve optimal therapeutic effect while avoiding subtherapeutic or toxic dosage levels.[3]

BIOLOGICAL PRINCIPLES OF THE PROCEDURE

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

1. Sample, anti-digoxin coated paramagnetic microparticles, assay diluent, and digoxigenin acridinium-labeled conjugate are combined to create a reaction mixture. The anti-digoxin coated microparticles bind to digoxin present in the sample and to the digoxigenin acridinium-labeled conjugate.
2. After washing, Pre-Trigger and Trigger Solutions are added to the reaction mixture.
3. The resulting chemiluminescent reaction is measured as relative light units (RLUs). There is an indirect relationship between the amount of digoxin in the sample and the RLUs detected by the ARCHITECT iSystem optics.

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

REAGENTS

Kit Contents

ARCHITECT iDigoxin 1P32

REF	1P32-27
Σ	100
MICROPARTICLES	1 x 6.6 mL
CONJUGATE	1 x 5.9 mL
ASSAY DILUENT	1 x 10.0 mL

MICROPARTICLES Anti-digoxin (mouse, monoclonal) coated microparticles in TRIS buffer with protein (bovine) stabilizer. Minimum concentration: 0.05% solids. Preservative: ProClin 300.

CONJUGATE Digoxigenin acridinium-labeled conjugate in citrate buffer. Minimum concentration: 1.0 ng/mL. Preservative: ProClin 300.

ASSAY DILUENT Assay Diluent containing goat serum with EDTA disodium. Preservatives: ProClin 300 and ProClin 950.

Other Reagents

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

TRIGGER SOLUTION ARCHITECT Trigger Solution containing 0.35 N sodium hydroxide.

WASH BUFFER ARCHITECT Wash Buffer containing phosphate buffered saline solution. Preservatives: antimicrobial agents.

Warnings and Precautions

- IVD
- For *In Vitro* Diagnostic Use

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. Biosafety Level 2 or other appropriate biosafety practices should be used for materials that contain or are suspected of containing infectious agents.[5-8]

The following warnings and precautions apply to: MICROPARTICLES and ASSAY DILUENT

WARNING	Contains methylisothiazolones.
H317	May cause an allergic skin reaction.
Prevention	
P261	Avoid breathing mist / vapors / 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.
P362+P364	Take off contaminated clothing and wash it before reuse.

Disposal	
P501	Dispose of contents / container in accordance with local regulations.

The following warnings and precautions apply to: CONJUGATE	
	
WARNING	Contains methylisothiazolones, 5-sulfosalicylic acid dihydrate, and citric acid monohydrate.
H317	May cause an allergic skin reaction.
H371	May cause damage to organs.
Prevention	
P260	Do not breathe mist / vapors / spray.
P280	Wear protective gloves / protective clothing / eye protection.
P264	Wash hands thoroughly after handling.
P272	Contaminated work clothing should not be allowed out of the workplace.
Response	
P302+P352	IF ON SKIN: Wash with plenty of water.
P333+P313	If skin irritation or rash occurs: Get medical advice / attention.
P308+P311	IF exposed or concerned: Call a POISON CENTER / doctor.
P362+P364	Take off contaminated clothing and wash it before reuse.
Disposal	
P501	Dispose of contents / container in accordance with local regulations.

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.

Reagent Handling

- Do not use reagent kits beyond the expiration date.
- **Do not pool reagents within a kit or between kits.**
- Before loading the 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 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 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.

Reagent Storage

When stored and handled as directed, reagents are stable until the expiration date.

	Storage Temperature	Maximum Storage Time	Additional Storage Instructions
Unopened/ Opened*	2-8˚C	Until expiration date	May be used immediately after removal from 2-8˚C storage. Store in upright position.
On board	System temperature	30 days	Discard after 30 days. For information on tracking onboard time, refer to the ARCHITECT System Operations Manual, Section 5.

* Reagents may be stored on or off the ARCHITECT iSystem. 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 iDigoxin assay file must be installed on the ARCHITECT iSystem with STAT protocol capability prior to 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.

Alternate Result Units

Edit assay parameter "Result concentration units" to select an alternate unit.

Conversion formula:

(Concentration in Default result unit) x (Conversion factor) = (Concentration in Alternate result unit)

Default result unit	Conversion factor	Alternate result unit
ng/mL	1.28	nmol/L

SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS

Specimen Types

Verified specimen types to be used with this assay:

Specimen Types	Collection Tubes
Human serum	Serum
Human plasma	Potassium EDTA Lithium heparin Sodium heparin

- Other specimen collection tubes, including gel separation tubes, have not been tested with this assay.
- Plasma samples from different anticoagulant tube types should not be used interchangeably for monitoring digoxin.
- Liquid anticoagulants may have a dilution effect resulting in lower concentrations for individual patient specimens.
- Performance has not been established for the use of cadaveric specimens or the use of body fluids other than human serum and plasma.
- The instrument 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 assay.

Specimen Conditions

- Do not use specimens with the following conditions:
  - heat-inactivated
  - grossly hemolyzed (> 750 mg/dL)
  - obvious microbial contamination
- 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.
- To prevent cross contamination, use of disposable pipettes or pipette tips is recommended.

Preparation for Analysis

- Follow the tube manufacturer's processing instructions for 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, centrifuge specimens before testing if
  - they contain fibrin, red blood cells, or other particulate matter,
  - they require repeat testing, or
  - they were frozen and thawed.
- Transfer clarified specimen to a sample cup or secondary tube for testing. For centrifuged specimens with a lipid layer, transfer only the clarified specimen and not the lipemic material.
- 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.

Specimen Storage

Specimen Type	Storage Temperature	Maximum Storage Time
Serum/Plasma	Room temperature	≤ 48 hours
	2-8°C	≤ 48 hours
	-20°C or colder	≤ 6 months[9]

Serum or plasma should be separated from the clot or red blood cells as soon after collection as possible.

Avoid more than 3 freeze/thaw cycles.

Specimen Shipping

- Package and label specimens in compliance with applicable state, federal, and international regulations covering the transport of clinical specimens and infectious substances.
- Do not exceed the storage limitations listed above.

PROCEDURE

Materials Provided

1P32 ARCHITECT iDigoxin Reagent Kit

Materials Required but not Provided

- ARCHITECT iDigoxin Assay file obtained from the ARCHITECT iSystem e-Assay CD-ROM found on www.abbottdiagnostics.com.
- 1P32-02 ARCHITECT iDigoxin Calibrators
- Commercial controls
- ARCHITECT Pre-Trigger Solution
- ARCHITECT Trigger Solution
- ARCHITECT Wash Buffer
- ARCHITECT Reaction Vessels
- ARCHITECT Sample Cups
- ARCHITECT Septum
- ARCHITECT Replacement Caps
- Pipettes or pipette tips (optional) to deliver the volumes specified on the patient or control order screen.

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

Assay Procedure

- Before loading the 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 local Abbott representative.**
  - Once the microparticles have been resuspended, place a septum on the bottle. For instructions about placing septums on bottles, refer to the **Reagent Handling** section of this package insert.
- Load the reagent kit on the ARCHITECT iSystem 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.
- Minimum sample volume is calculated by the system and is printed on the Orderlist report. To minimize the effects of evaporation, verify that adequate sample cup volume is present prior to running the test.
- Maximum number of replicates sampled from the same sample cup: 10
  - Priority:

    Sample volume for the first test: 100 µL

    Sample volume for each additional test from the same sample cup: 50 µL
- ≤ 3 hours on board:

  Sample volume for the first test: 150 µL

  Sample volume for each additional test from the same sample cup: 50 µL

- To minimize the effects of evaporation all samples (patient specimens, calibrators and controls) must be tested within 3 hours of being placed on board the ARCHITECT iSystem.
  - If using primary or aliquot tubes, use the sample gauge to ensure that sufficient patient specimen is present.
- Prepare ARCHITECT iDigoxin Calibrators and controls.
  - Mix calibrators by gentle inversion before use.
  - Hold bottles **vertically** and dispense recommended volumes into each respective sample cup.
- Recommended volumes:
  for each calibrator: 5 drops
- Dispense 150 µL of each control 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 digoxin value exceeding 4.00 ng/mL are flagged with the code ">4.00 ng/mL"and may be diluted using the Manual Dilution Procedure.

Manual Dilution Procedure

Suggested dilution: 1:10

1. Add 20 µL of the patient specimen to 180 µL of Calibrator A.
2. 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 and report the result. The result should be > 0.30 ng/mL before the dilution factor is applied.

For detailed information on ordering dilutions, refer to the ARCHITECT System Operations Manual, Section 5.

Calibration

- Test Calibrators A-F in duplicate. The calibrators should be priority loaded.
  A single sample of each control level must be tested to evaluate the assay calibration. Ensure that assay control values are within the ranges specified in the respective control package insert.
- Calibration Range: 0.0 to 4.0 ng/mL.
- Once an ARCHITECT iDigoxin 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 or
  - 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 iDigoxin assay is that a single sample of each control level be 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.

Additional controls may be tested in accordance with local, state, and/or federal regulations or accreditation requirements and your laboratory's quality control policy.

The control values must be within the acceptable ranges specified in the control package insert. If a control is out of its specified range, the associated sample results are invalid and the samples must be retested. Recalibration may be indicated.

Verification of Assay Claims

For protocols to verify package insert claims, refer to the ARCHITECT System Operations Manual, Appendix B.

The ARCHITECT iDigoxin assay belongs to method group 2.

RESULTS

Calculation

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

Flags

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

Measurement Range (Reportable Range)

The measurement range for the ARCHITECT iDigoxin assay is 0.3 to 4.0 ng/mL.

LIMITATIONS OF THE PROCEDURE

- For diagnostic purposes, results should be used in conjunction with other data; e.g., symptoms, results of other tests, clinical impressions, etc.
- If the ARCHITECT iDigoxin results are inconsistent with clinical evidence, additional testing is recommended.
- Plasma samples from different anticoagulant tube types should not be used interchangeably for monitoring digoxin.
- Specimens from patients who have received preparations of mouse monoclonal antibodies for diagnosis or therapy may contain human anti-mouse antibodies (HAMA). Such specimens may show either falsely elevated or depressed values when tested with assay kits such as ARCHITECT iDigoxin that employ mouse monoclonal antibodies.[10-12]
- Some immunoassays for digoxin may cross-react with metabolites, which can lead to a positive bias in patient results. Refer to the **SPECIFIC PERFORMANCE CHARACTERISTICS**, **Specificity** section of this package insert for estimates of cross-reactivity of ARCHITECT iDigoxin to some metabolites of digoxin.
- Digoxin values for specimens from patients who have received DIGIBIND or DIGIFAB therapy may be impacted. See the **SPECIFIC PERFORMANCE CHARACTERISTICS**, **Interfering Substances** section.
- Heterophilic antibodies in human serum can react with reagent immunoglobulins, interfering with *in vitro* immunoassays.[12] The presence of heterophilic antibodies in a patient specimen may cause anomalous values to 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

The ARCHITECT iDigoxin assay accurately quantitates digoxin concentrations in human serum or plasma at concentrations up to 4.0 ng/mL. Numerous studies have shown a relationship between serum levels of digoxin and its concentration in myocardial and other tissues. In a study in which serum digoxin levels were determined by radioimmunoassay, optimum therapeutic effects usually are observed when serum levels are in the range from 0.8 to 2.0 ng/mL[13], although some clinical benefit may be realized at serum or plasma concentrations below 0.8 ng/mL.[14] The risk of toxicity increases at serum or plasma levels above 2.0 ng/mL.[14] Symptoms of digoxin toxicity may include gastrointestinal disturbances such as anorexia, nausea, vomiting and diarrhea, central nervous system disturbances manifested by blurred or yellow vision, headache, weakness, dizziness, apathy, and confusion, and cardiac rhythm disturbances and tachycardia.[14] There is some evidence that children may tolerate slightly higher serum or plasma concentrations than adults.[14] It is important to note that the distinction between adequate digitalization and toxicity in patients cannot be made on the basis of digoxin concentrations alone. Most studies show a significant overlap between the toxic and nontoxic groups. Additional factors to consider when evaluating the correct therapeutic dosage for each patient are lean body weight, age, renal function, concomitant disease states, concurrent medications, and other clinical factors.[14] Refer to the drug manufacturer's package insert. Each laboratory should establish its own therapeutic (reference) range for digoxin.

SPECIFIC PERFORMANCE CHARACTERISTICS

Performance was evaluated on the ARCHITECT i2000SR System.

Precision

The ARCHITECT iDigoxin assay is designed to have an assay precision of ≤ 10% total CV.

A study was performed with guidance from the Clinical and Laboratory Standards Institute (CLSI) Document EP5-A2.[15] Abbott Immunoassay-MCC (Liquid) (Levels 1, 2, and 3) and three human serum panels were assayed in replicates of two at two separate times per day for 20 days using two lots of reagents on two instruments. 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 (ng/mL)	Within Run SD	Within Run %CV	Total SD	Total %CV
Level 1	1	1	80	0.81	0.020	2.5	0.024	3.0
	2	2	80	0.82	0.020	2.4	0.027	3.3
Level 2	1	1	80	1.72	0.032	1.9	0.053	3.1
	2	2	80	1.73	0.030	1.7	0.040	2.3
Level 3	1	1	80	2.88	0.069	2.4	0.075	2.6
	2	2	80	2.89	0.055	1.9	0.059	2.0
Panel 1	1	1	80	0.66	0.024	3.6	0.029	4.4
	2	2	80	0.68	0.017	2.5	0.025	3.7
Panel 2	1	1	80	1.70	0.039	2.3	0.059	3.5
	2	2	80	1.72	0.026	1.5	0.035	2.0
Panel 3	1	1	80	3.53	0.064	1.8	0.118	3.3
	2	2	80	3.57	0.056	1.6	0.087	2.4

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

Recovery

The ARCHITECT iDigoxin assay is designed to have a grand mean recovery of 100 ± 10%.

A study was performed on five serum samples containing low levels of digoxin and each sample was spiked with additional digoxin at concentrations of 0.0, 0.5, 1.0, 2.0, and 3.0 ng/mL. The concentration of digoxin was determined using the ARCHITECT iDigoxin assay and the resulting percent recovery was calculated. The individual percent recovery of the ARCHITECT iDigoxin assay for serum ranged from 96.0% to 104.3%. The mean percent recovery of the ARCHITECT iDigoxin assay for serum ranged from 99.9% to 102.9% with a grand mean for serum of 101.8%.*

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

Dilution Linearity

The ARCHITECT iDigoxin assay is designed to have a mean deviation from linearity of ± 10% for concentrations over 1.0 ng/mL or ± 0.1 ng/mL at concentrations less than 1.0 ng/mL.

A dilution linearity study was performed by diluting five serum samples with ARCHITECT iDigoxin Calibrator A. The concentration of digoxin was determined using the ARCHITECT iDigoxin assay and the resulting percent deviation from linearity or concentration difference was calculated. For diluted samples reading above 1.0 ng/mL, the percent deviation from linearity ranged from -4.0 to 8.8% with a mean percent deviation from linearity of 2.0%. For diluted samples below 1.0 ng/mL, the concentrations were within 0.1 ng/mL of the expected results.* The linear range of the assay is 0.3 to 4.0 ng/mL.

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

Sensitivity

The ARCHITECT iDigoxin assay is designed to have a Limit of Detection of ≤ 0.3 ng/mL.

Limit of Blank (LoB) and Limit of Detection (LoD)

The LoB and LoD of the ARCHITECT iDigoxin assay were determined with guidance from CLSI Document EP17-A[16] using proportions of false positives (α) less than 5% and false negatives (β) less than 5%. These determinations were performed using one blank (60 replicates) and four low level digoxin samples (15 replicates each); LoB = 0.07 ng/mL and LoD = 0.09 ng/mL.*

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

Functional Sensitivity

In order to perform the Functional Sensitivity study, a series of seven samples ranging from 0.05 to 0.5 ng/mL were prepared by diluting Calibrator B with Calibrator A. These samples were tested in replicates of ten two times per day for five days using one reagent and calibrator lot for a total of 100 replicates per panel. The total % CVs were calculated and plotted against the mean concentration. A reciprocal curve was fitted through the data, and the functional sensitivity value was calculated as the concentration corresponding to the 20% CV level of the fitted curve. At the upper 95% confidence limit, the lowest ARCHITECT iDigoxin assay value exhibiting a 20% CV was calculated to be 0.1 ng/mL, which is below the reportable range of the ARCHITECT iDigoxin assay.*

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

Specificity

Cross-reactivity was tested for the major digoxin active metabolites (digoxigenin bis-digitoxoside, digoxigenin mono-digitoxoside, digoxigenin), related compounds (acetyldigoxin, digitoxin, digitoxigenin, ouabain, deslanoside, lanatoside C, proscillaridin) and other therapeutic agents that may be coadministered to determine whether these compounds affect the quantitation of digoxin concentrations using the ARCHITECT iDigoxin assay. The compounds were spiked into a serum pool (control) containing no digoxin and into two therapeutic levels (approximately 0.8 ng/mL and 2.0 ng/mL) of digoxin. The samples were assayed and the digoxin concentrations of the spiked samples were compared to the control serum. The data are summarized in the following table.*

Test Compound	Test Compound Conc. (ng/mL)	Digoxin Concentration (ng/mL) 0.0 Conc. Diff.[a]	0.8 Conc. Diff.[a]	0.8 Cross-React. (%)[b]	2.0 Conc. Diff.[a]	2.0 Cross-React. (%)[b]
Digoxigenin bis-digitoxoside	1.30	2.03	1.86	143.1	—[c]	—[c]
Digoxigenin mono-digitoxoside	1.00	1.49	0.69	69.0	0.96	96.0
Digoxigenin	0.80	0.15	0.02	2.5	-0.05	-6.3
Acetyldigoxin	2.50	2.70	2.34	93.6	—[c]	—[c]
Digitoxin	25	0.04	0.07	0.3	-0.09	-0.4
Digitoxigenin	15	0.00	0.04	0.3	-0.08	-0.5
Ouabain	860	1.38	0.28	0.0	0.22	0.0
Deslanoside	2.25	2.03	1.65	73.3	1.66	73.8
Lanatoside C	1.55	1.98	1.14	73.5	0.96	61.9
Proscillaridin	340	0.21	0.26	0.1	0.08	0.0
Spironolactone	500	0.35	0.04	0.0	0.21	0.0
Canrenone	1000	0.00	0.01	0.0	0.05	0.0
Canrenoic Acid	1000	0.36	0.08	0.0	-0.04	0.0
Hydrocortisone	2000	0.12	0.02	0.0	0.01	0.0
Methylprednisolone	7000	0.02	0.08	0.0	-0.05	0.0
Prednisolone	1000	0.15	0.02	0.0	0.37	0.0
Dexamethasone	5000	0.36	0.19	0.0	0.10	0.0
Progesterone	250	0.05	0.06	0.0	-0.12	0.0
Amiloride	50	0.00	0.36	0.7	-0.04	-0.1
Amrinone	7000	0.15	-0.11	0.0	0.11	0.0
Clorazepate	1500	0.00	0.04	0.0	-0.09	0.0
Nabumetone	37,000	0.00	0.12	0.0	-0.04	0.0
Phenytoin	80,000	0.00	0.16	0.0	-0.03	0.0
Triamterene	500	0.00	-0.25	-0.1	-0.03	0.0

[a] Concentration Difference

= Measured value of sample spiked with test compound – Measured value of control

[b] $\% \text{ Cross-reactivity} = 100 * \frac{\text{Measured value of sample spiked with test compound} - \text{Measured value of control}}{\text{Test compound concentration}}$

[c] Not Determined. The concentration value measured is outside of the assay range (> 4.00 ng/mL).

It has been reported in the literature that one in ten patients converts 40% or more of oral digoxin to an inactive reduction product (dihydrodigoxin) via bacteria in the gut.[14] There is little or no cross-reactivity reported for dihydrodigoxin.[17]

A case report by Steimer et al.[18] showed that a negative bias in the determination of digoxin results may be observed when the aldosterone inhibitors spironolactone or canrenone are present in serum. The aldosterone inhibitor results show no significant interference with the ARCHITECT iDigoxin assay.

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

Interfering Substances

Potential interference in the ARCHITECT iDigoxin assay from the following compounds is designed to have a mean recovery of 100 ± 10% of the control results at the levels indicated. A study based on guidance from the CLSI Document EP7-A2[19] was performed for the ARCHITECT iDigoxin assay. Five serum specimens were targeted at digoxin concentrations of 0.5, 0.8, 1.5, 2.0 and 3.0 ng/mL and supplemented with the following potentially interfering compounds except for low and high protein. The low and high protein samples were prepared before supplementing with digoxin at the above target concentrations. The data are summarized in the following table.*

Potentially Interfering Compound	Interferent Concentration	Percent Recovery Range (Individual)
Bilirubin	20 mg/dL	98.9 – 103.1
Hemoglobin	750 mg/dL	100.0 – 103.2
Triglycerides	2500 mg/dL	98.4 – 105.6
HAMA	1000 ng/mL	98.1 – 101.6
Rheumatoid Factor	500 IU/mL	97.1 – 102.5
Low Protein	3 g/dL	94.3 – 106.8
High Protein	12 g/dL	97.6 – 109.8

The mean percent recovery in this study ranged from 99.1% to 104.8%.*

The sera from patients in specific patient populations (i.e., patients with renal and/or hepatic failure, newborn infants, and pregnant women) have been reported to contain an unidentified component that gives positive results for digoxin with a number of immunoassays.[20-26] This component has been called digoxin-like immunoreactive factor (DLIF) or substance (DLIS). The presence of DLIF in a sample can result in falsely elevated digoxin assay results. The amount of DLIF in these patient samples is extremely variable, but in some cases these levels have been shown to approach concentrations that are in the therapeutic range of digoxin.[21, 22, 24]

As with any assay employing mouse antibodies, the possibility exists for interference by HAMA in the sample, which could falsely elevate or depress results.

The manufacturer of Digoxin Immune Fab has stated that no immunoassay technique is suitable for quantitating digoxin in serum from patients on antibody fragment therapy. According to the manufacturer's insert, DIGIBIND will interfere with digitalis immunoassay measurements.[27, 28]

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

Method Comparison

The ARCHITECT iDigoxin assay is designed to have a slope of 1.0 ± 0.1 and a correlation coefficient (r) of ≥ 0.95 for specimens when compared to MULTIGENT Digoxin. A study was performed using serum and plasma specimens. The data are summarized in the following table.*

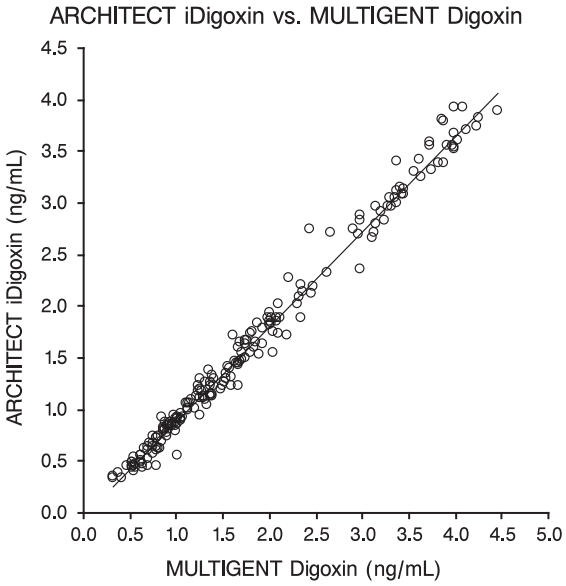
ARCHITECT iDigoxin vs. MULTIGENT Digoxin				
Regression Method	Number of Observations	Slope (95% CI[a])	Intercept (95% CI[a])	Correlation Coefficient
Passing-Bablok[b]	200	0.921 (0.906 to 0.937)	-0.040 (-0.061 to -0.018)	0.993

Specimen Range (ARCHITECT) = 0.34 ng/mL to 3.93 ng/mL

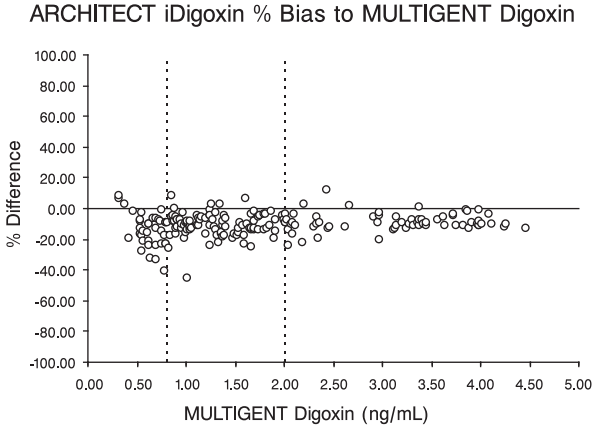
Specimen Range (MULTIGENT) = 0.32 ng/mL to 4.46 ng/mL

[a] CI = Confidence Interval

[b] A linear regression method with no special assumptions regarding the distribution of the samples and measurement errors.[29]



A bias analysis of ARCHITECT iDigoxin vs. MULTIGENT Digoxin was performed on the same 200 specimens in the range of 0.34 ng/mL to 3.93 ng/mL and 0.32 ng/mL to 4.46 ng/mL, respectively. The following representative data are provided to aid in understanding the difference between the two assays. The average bias exhibited by ARCHITECT vs. MULTIGENT in this study was -10.78%. The 95% confidence interval of that average bias is -26.72% to 5.16%. Within the typical therapeutic range of digoxin therapy (0.8 to 2.0 ng/mL, as read by MULTIGENT Digoxin), the average bias was -10.77% with a 95% confidence interval of -25.61% to 4.06%. Results of the study are summarized in the following graph.* The vertical lines depict the typical therapeutic range of digoxin therapy.



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

BIBLIOGRAPHY

1. Brunton LL, ed. Goodman & Gilman's *The Pharmacological Basis of Therapeutics.* [Internet]. 11th ed. USA: McGraw-Hill; 2006. Chapter 33: Rocco TP, Fang JC. Pharmacotherapy of congestive heart failure;[cited 2008 June 30]; [about 37 p]. Available from: http://www.accessmedicine.com/resourceTOC.aspx?resourceID=28. Accessed June 30, 2008.
2. Brunton LL, ed. Goodman & Gilman's *The Pharmacological Basis of Therapeutics.* [Internet]. 11th ed. USA: McGraw-Hill; 2006. Chapter 34: Roden DM. Antiarrhythmic drugs; [cited 2008 June 30]; [about 37 p].Available from: http://www.accessmedicine.com/resourceTOC.aspx?resourceID=28. Accessed June 30, 2008.
3. Valdes R, Jortani SA, Gheorghiade M. Standards of laboratory practice: cardiac drug monitoring. *Clin Chem.* 1998;44:1096-1109.
4. Gheorghiade M, Adams KF Jr, Colucci WS. Digoxin in the management of cardiovascular disorders. *Circulation.* 2004;109:2959-2964.
5. US Department of Labor, Occupational Safety and Health Administration, 29 CFR Part 1910.1030, Bloodborne pathogens.
6. US Department of Health and Human Services. *Biosafety in Microbiological and Biomedical Laboratories*. 5th ed. Washington, DC: US Government Printing Office; December 2009.
7. World Health Organization. *Laboratory Biosafety Manual*. 3rd ed. Geneva: World Health Organization; 2004.
8. Clinical and Laboratory Standards Institute (CLSI). *Protection of Laboratory Workers From Occupationally Acquired Infections; Approved Guideline—Fourth Edition.* CLSI Document M29-A4. Wayne, PA: CLSI; 2014.
9. Guder WG, Narayanan S, Wisser H, Zawta B. Digoxin. In: *Samples: From the Patient to the Laboratory.* 3rd ed. Germany: Wiley-VCH GmbH & Co. KGaA 2003; Annex [CD-ROM]:140.
10. Primus FJ, Kelley EA, Hansen HJ, et al. "Sandwich"-type immunoassay of carcinoembryonic antigen in patients receiving murine monoclonal antibodies for diagnosis and therapy. *Clin Chem* 1988;34(2):261-264.
11. Schroff RW, Foon KA, Beatty SM, et al. Human anti-murine immunoglobulin responses in patients receiving monoclonal antibody therapy. *Cancer Res* 1985;45(2):879-885.
12. Boscato LM, Stuart MC. Heterophilic antibodies: a problem for all immunoassays. *Clin Chem* 1988;34(1):27-33.
13. Smith TW, Butler VP, Haber E. Determination of therapeutic and toxic serum digoxin concentrations by radioimmunoassay. *New Engl J Med.* 1969;281:1212-1216.
14. Lanoxin. In: *Physicians' Desk Reference.* 62nd ed. Montvale, NJ: Thompson Healthcare Inc.; 2007:1498-1501.
15. National Committee for Clinical Laboratory Standards (NCCLS). *Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline—Second Edition.* NCCLS Document EP5-A2. Wayne, PA: NCCLS; 2004.
16. National Committee for Clinical Laboratory Standards (NCCLS). *Protocols for Determination of Limits of Detection and Limits of Quantitation; Approved Guideline.* NCCLS Document EP17-A. Wayne, PA: NCCLS; 2004.
17. Miller JJ, Straub RW Jr, Valdes R Jr. Digoxin immunoassay with cross-reactivity of digoxin metabolites proportional to their biological activity. *Clin Chem.* 1994;40:1898-1903.
18. Steimer W, Muller C, Eber B, et al. Intoxication due to negative canrenone interference in digoxin drug monitoring. *Lancet.* 1999;354:1176–1177.
19. Clinical and Laboratory Standards Institute (CLSI). *Interference Testing in Clinical Chemistry; Approved Guideline—Second Edition.* CLSI Document EP7-A2. Wayne, PA: CLSI; 2005.
20. Yatscoff RW, Desjardins PR, Dalton JG. Digoxin-like immunoreactivity in the serum of neonates and uremic patients, as measured in the Abbott TDx. *Clin Chem.* 1984;30:588.
21. Greenway DC, Nanji AA. Falsely increased results for digoxin in sera from patients with liver disease: ten immunoassay kits compared. *Clin Chem.* 1985;31:1078–1079.
22. Rosenkranz B, Frolich JC. Falsely elevated digoxin concentrations in patients with liver disease. *Ther Drug Monit.* 1985;7:202–206.
23. Pudek MR, Seccombe DW, Jacobson BE, et al. Seven different digoxin immunoassay kits compared with respect to interference by a digoxin-like immunoreactive substance in serum from premature and full-term infants. *Clin Chem.* 1983;29:1972–1974.
24. Hicks JM, Brett EM. Falsely increased digoxin concentrations in samples from neonates and infants. *Ther Drug Monit.* 1984;6:461–464.
25. Soldin SJ, Papanastasiou-Diamandi A, Heyes J, et al. Are immunoassays for digoxin reliable? *Clin Biochem.* 1984;17:317–320.
26. Valdes R Jr. Endogenous digoxin-like immunoreactive factors: impact on digoxin measurements and potential physiological implications. *Clin Chem.* 1985;31:1525–1532.
27. DIGIBIND. In: *Physician's Desk Reference.* 62nd ed. Montvale, NJ: Thomson Healthcare Inc. 2007;1411-1413.
28. Rainey PM. Effects of digoxin immune Fab (ovine) on digoxin immunoassays. *Am J Clin Pathol.* 1989;92:779–786.
29. Passing H, Bablok W. A new biometrical procedure for testing the equality of measurements from two different analytical methods. Application of linear regression procedures for method comparison studies in clinical chemistry, Part I. *J Clin Chem Clin Biochem* 1983;21(11):709–720.

Key to Symbols

	Consult instructions for use
	Manufacturer
	Sufficient for
	Temperature limitation
	Use by/Expiration date
ASSAY DILUENT	Assay Diluent
CONJUGATE	Conjugate
CONTROL NO.	Control Number
EC REP	Authorized Representative in the European Community
INFORMATION FOR USA ONLY	Information needed for United States of America only
IVD	*In Vitro* Diagnostic Medical Device
LOT	Lot Number
MICROPARTICLES	Microparticles
PRE-TRIGGER SOLUTION	Pre-Trigger Solution
PRODUCED FOR ABBOTT BY	Produced for Abbott by
PRODUCT OF SPAIN	Product of Spain
REACTION VESSELS	Reaction Vessels
REAGENT LOT	Reagent Lot
REF	List Number
REPLACEMENT CAPS	Replacement Caps
SAMPLE CUPS	Sample Cups
SEPTUM	Septum
SN	Serial number
TRIGGER SOLUTION	Trigger Solution
WASH BUFFER	Wash Buffer

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