Total Bilirubin2

FOR USE WITH

ARCHITECT

en
TBili2
04T09
H06717R03
B4T090

Read Highlighted Changes: Revised April 2025.

REF 04T0920

REF 04T0930

Instructions must be carefully followed. Reliability of assay results cannot be guaranteed if there are any deviations from these instructions.

For laboratory professional use only.

NAME

Total Bilirubin2 (also referred to as TBili2)

INTENDED USE

The Total Bilirubin2 assay is used for the quantitation of total bilirubin in human serum or plasma, of adults and neonates, on the ARCHITECT c Systems.

Measurement of total bilirubin, an organic compound formed during the normal and abnormal destruction of red blood cells, is used in the diagnosis and treatment of liver, hemolytic, hematological, and metabolic disorders, including hepatitis and disorders of the biliary tract. In newborn infants, the Total Bilirubin2 assay is intended to measure the levels of total bilirubin (conjugated and unconjugated) in serum or plasma to aid in the diagnosis and management of neonatal jaundice and hemolytic disease of the newborn.

SUMMARY AND EXPLANATION OF THE TEST

Bilirubin is a degradation product of hemoglobin. Bilirubin bound to albumin is insoluble in water and is known as unconjugated (indirect) bilirubin. In the liver, unconjugated bilirubin is coupled with glucuronide; this form is called conjugated (direct) bilirubin. It is water soluble and is mostly excreted in bile.[1]

The sum of direct and indirect bilirubin is called total bilirubin and the indirect fraction of the total usually makes up to approximately 85%.[1]

In cases of hyperbilirubinemia, bile pigment is deposited in the skin, sclera, and mucous membranes and so the patient has yellowish color; this condition is called jaundice or icterus. In newborns or in people with familial hyperbilirubinemia the presence of jaundice and elevation of total bilirubin may indicate inherited metabolic disorders (e.g., Gilbert, Crigler-Najjar, Lucey-Driscoll, Dubin-Johnson, and Rotor syndromes).[2]

Fractionation of total bilirubin into conjugated and unconjugated may help in the diagnosis of hyperbilirubinemia. For example, conjugated bilirubin is increased in cases of cholestasis caused by several liver diseases (e.g., hepatitis, hepatic obstruction, and cirrhosis), while a high level of unconjugated bilirubin may indicate a hemolytic disorder.[2] Inherited metabolic disorders may also have differences in conjugated and unconjugated fractions: increased conjugated bilirubin suggests Dubin-Johnson or Rotor syndromes, while unconjugated bilirubin is prevalent in Gilbert, Crigler-Najjar, or Lucey-Driscoll syndromes.[2]

The total bilirubin test is used as an aid in the differential diagnosis and management of liver diseases, and neonatal jaundice, as well as hemolytic, and inherited metabolic diseases.[2]

PRINCIPLES OF THE PROCEDURE

The Total Bilirubin2 assay is an automated clinical chemistry assay.

Total (conjugated and unconjugated) bilirubin couples with a diazo reagent in the presence of a surfactant to form azobilirubin. The diazo reaction is accelerated by the addition of surfactant as a solubilizing agent. The increase in absorbance at 548 nm due to azobilirubin is directly proportional to the total bilirubin concentration.

Methodology: Diazonium salt

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

REAGENTS

Kit Contents

Total Bilirubin2 Reagent Kit 04T09

NOTE: Some kit sizes may not be available. Please contact your local distributor.

Volumes (mL) listed in the following table indicate the volume per cartridge.

REF	04T0920	04T0930
Tests per cartridge set	225	450
Number of cartridge sets per kit	4	8
Tests per kit	900	3600
R1	44.5 mL	85.9 mL
R2	14.5 mL	26.7 mL

R1 Active ingredient: Brij L23 (233.333 mL/L).

R2 Active ingredients: 2,4-dichlorobenzenediazonium 1,5-naphthalenedisulfonate hydrate (1845.000 mg/L) and Brij L23 (100.000 mL/L).

Warnings and Precautions

- IVD
- For *In Vitro* Diagnostic Use
- Rx ONLY

Safety Precautions

CAUTION: This product requires the handling of human specimens. It is recommended that all human-sourced materials and all consumables contaminated with potentially infectious materials be considered potentially infectious and handled in accordance with the OSHA Standard on Bloodborne Pathogens. Biosafety Level 2 or other appropriate regional, national, and institutional biosafety practices should be used for materials that contain, are suspected of containing, or are contaminated with infectious agents.[3-6]

The following warnings and precautions apply to: R1	
	
DANGER	Contains sodium metaborate tetrahydrate, polyoxyethylene lauryl ether, and phosphoric acid.
H360*	May damage fertility or the unborn child.
H360FD**	May damage fertility. May damage the unborn child.
H290	May be corrosive to metals.
H314	Causes severe skin burns and eye damage.
H402*	Harmful to aquatic life.
H412	Harmful to aquatic life with long lasting effects.
Prevention	
P201	Obtain special instructions before use.
P234	Keep only in original container.
P260	Do not breathe mist / vapors / spray.
P264	Wash hands thoroughly after handling.
P273	Avoid release to the environment.

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P280	Wear protective gloves / protective clothing / eye protection.
Response	
P301+P330+P331	IF SWALLOWED: Rinse mouth. Do NOT induce vomiting.
P303+P361+P353	IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water / shower.
P305+P351+P338	IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P308+P313	IF exposed or concerned: Get medical advice / attention.
P310	Immediately call a POISON CENTER or doctor / physician.
P390	Absorb spillage to prevent material damage.
Disposal	
P501	Dispose of contents / container in accordance with local regulations.

* Not applicable where regulation EC 1272/2008 (CLP) has been implemented.

** Applicable only where regulation EC 1272/2008 (CLP) has been implemented.

The following warnings and precautions apply to: R2

DANGER	Contains polyoxyethylene lauryl ether and hydrochloric acid.
H290	May be corrosive to metals.
H314	Causes severe skin burns and eye damage.
H402*	Harmful to aquatic life.
H412	Harmful to aquatic life with long lasting effects.
Prevention	
P234	Keep only in original container.
P260	Do not breathe mist / vapors / spray.
P264	Wash hands thoroughly after handling.
P273	Avoid release to the environment.
P280	Wear protective gloves / protective clothing / eye protection.
Response	
P301+P330+P331	IF SWALLOWED: Rinse mouth. Do NOT induce vomiting.
P303+P361+P353	IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water / shower.
P305+P351+P338	IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P310	Immediately call a POISON CENTER or doctor / physician.
P390	Absorb spillage to prevent material damage.
Disposal	
P501	Dispose of contents / container in accordance with local regulations.

* Not applicable where regulation EC 1272/2008 (CLP) has been implemented.

Follow local chemical disposal regulations based on your location along with recommendations and content in the Safety Data Sheet to determine the safe disposal of this product.

For the most current hazard information, see the product Safety Data Sheet.

Safety Data Sheets are available at www.corelaboratory.abbott 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 pool reagents within a kit or between kits.
- Do not use components from one lot with components from another lot.
- Do not reuse containers, caps or plugs due to the risk of contamination and the potential to compromise reagent performance.
- When either the R1 or R2 reagent cartridge becomes empty, replace both cartridges.
- Upon receipt, place reagent cartridges in an upright position for 6 hours before use to allow bubbles that may have formed to dissipate.
- If a reagent cartridge is dropped, place in an upright position for 2 hours before use to allow bubbles that may have formed to dissipate.
- Reagents are susceptible to the formation of foam and bubbles. Bubbles may interfere with the detection of the reagent level in the cartridge and cause insufficient reagent aspiration that may adversely affect results.

For a detailed discussion of reagent handling precautions during system operation, refer to the ARCHITECT System Operations Manual, Section 7.

Reagent Storage

	Storage Temperature	Maximum Storage Time	Additional Storage Instructions
Unopened	2 to 8°C	Until expiration date	Store in upright position.
Onboard	System Temperature	30 days	
Opened	2 to 8°C	Until expiration date	Store in upright position.

Reagents may be stored on or off the ARCHITECT c System. If reagents are removed from the system, store at 2 to 8°C (with replacement caps) in their original boxes.

For information on unloading reagents, refer to the ARCHITECT System Operations Manual, Section 5.

Indications of Reagent Deterioration

Deterioration of the reagents may be indicated when a calibration error occurs or a control value is out of the specified range. 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 Total Bilirubin2 assay file must be installed on the ARCHITECT c System prior to performing the assay.

Installation of all the required SmartWash updates on either the MULTIGENT Assay Disk Version 9.00 (or higher) or the Special Chemistry Assay Disk Version 7.00 (or higher) must be completed prior to performing the assay. See below for impacted assays:

				VERSION	
Assay Name	Short Name	REF	Assay Number	Conventional Units / Alternate Units	SI Units / Alternate Units
D-Dimer	dDim	7K02	2904	6	6

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.

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Alternate Result Units

Conversion formula:

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

Default Result Unit	Conversion Factor	Alternate Result Unit
mg/dL	17.1	µmol/L

SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS

Specimen Types

The specimen types listed below were verified for use with this assay.

Other specimen types and collection tube types have not been verified with this assay.

Specimen Types	Collection Tubes	Special Conditions
Serum	Serum	Protect from light.[7]
	Serum separator	
Plasma	Dipotassium EDTA	Protect from light.[7]
	Lithium heparin	
	Lithium heparin separator	
	Sodium heparin	

- Liquid anticoagulants may have a dilution effect resulting in lower concentration values for individual specimens.

The instrument does not provide the capability to verify specimen types. It is the responsibility of the operator to verify that the correct specimen types are used in the assay.

Specimen Conditions

- Do not use:
  - heat-inactivated specimens
  - pooled specimens
  - grossly hemolyzed specimens
  - specimens with obvious microbial contamination
  - specimens with fungal growth
- 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.
- Specimens should be free of bubbles. Remove bubbles with an applicator stick before analysis. Use a new applicator stick for each specimen to prevent cross contamination.

To ensure consistency in results, recentrifuge specimens prior to testing if

- they contain fibrin, red blood cells, or other particulate matter.

NOTE: If fibrin, red blood cells, or other particulate matter are observed, mix by low-speed vortex or by inverting 10 times prior to recentrifugation.

Prepare frozen specimens as follows:

- Frozen specimens must be completely thawed before mixing.
- Mix thawed specimens thoroughly by low-speed vortex or by inverting 10 times.
- Visually inspect the specimens. If layering or stratification is observed, mix until specimens are visibly homogeneous.
- If specimens are not mixed thoroughly, inconsistent results may be obtained.
- Recentrifuge specimens.

Recentrifugation of Specimens

- Transfer specimens to a centrifuge tube and centrifuge.
- 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.

Specimen Storage

Specimen Type	Temperature	Collection Tubes	Maximum Storage Time
Serum/ Plasma	Room temperature (20 to 25°C)	Serum Dipotassium EDTA Lithium heparin Sodium heparin	8 hours[8]
		Serum separator Lithium heparin separator	24 hours[9]
	2 to 8°C	Serum Serum separator Dipotassium EDTA Lithium heparin Lithium heparin separator Sodium heparin	7 days[10]
	-20°C	Serum Serum separator Dipotassium EDTA Lithium heparin Lithium heparin separator Sodium heparin	3 months[11]

Specimens should be protected from light as bilirubin is photolabile.[7]

Avoid multiple freeze/thaw cycles.[11]

It is the responsibility of the individual laboratory to determine specific specimen stability criteria for their laboratory per their laboratory workflow.

For additional information on sample handling and processing, refer to CLSI GP44-A4.[12] The storage information provided here is based on references or data maintained by the manufacturer.

Each laboratory may establish a range around -20°C from either the freezer manufacturer's specifications or your laboratory standard operating procedure(s) for specimen storage.

Stored specimens must be inspected for particulates. If present, mix with a low-speed vortex or by inversion and centrifuge the specimen to remove particulates prior to testing.

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

04T09 Total Bilirubin2 Reagent Kit

Materials Required but not Provided

- Total Bilirubin2 assay file found on www.corelaboratory.abbott
- 04V1501 Consolidated Chemistry Calibrator
- Controls containing total bilirubin
- Saline (0.85% to 0.90% NaCl) for specimen dilution

For information on materials required for operation of the instrument, refer to the ARCHITECT System Operations Manual, Section 1.

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

Assay Procedure

For a detailed description of how to run an assay, refer to the ARCHITECT System Operations Manual, Section 5.

- If using primary or aliquot tubes, refer to the ARCHITECT System Operations Manual, Section 5 to ensure sufficient specimen is present.
- Minimum sample cup volume is calculated by the system and printed on the Order List report. To minimize the effects of evaporation, verify adequate sample cup volume is present prior to running the test.
- Minimum sample volume requirements:
  - Sample volume for single test: 2.6 µL.

NOTE: This amount does not include the dead volume plus the additional over-aspiration volume. For total sample volume requirements, refer to the ARCHITECT System Operations Manual, Section 5.

- Refer to the Consolidated Chemistry Calibrator package insert REF 04V1501 and/or commercially available control material package insert for preparation and usage.
- For general operating procedures, refer to the ARCHITECT System Operations Manual, Section 5.
- 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.

Sample Dilution Procedures

The standard dilution factor, applied automatically by the system software to all results for the Total Bilirubin2 assay, is 1:1.97.

Samples with a total bilirubin value exceeding 25.0 mg/dL (427.5 µmol/L) are flagged with the code "> 25.0 mg/dL" ("> 427.5 µmol/L") and may be diluted with either the Automated Dilution Protocol or the Manual Dilution Procedure.

Automated Dilution Protocol

The automated dilution factor for the Total Bilirubin2 assay is 1:9.87.

The system performs a dilution of the sample, relative to the standard dilution, and automatically calculates the concentration by multiplying the result by the dilution factor.

Dilution Name	Dilution Factor
STANDARD	1:1.97
1:5	1:9.87

For details on configuring automated dilutions, refer to the ARCHITECT System Operations Manual, Section 2.

Manual Dilution Procedure

Specimens with a total bilirubin value exceeding 25.0 mg/dL (427.5 µmol/L) can be manually diluted using a 1:5 dilution.

Dilute the sample with saline (0.85% to 0.90% NaCl).

The operator must enter the manual dilution factor (5) in the Patient or Control order screen. The system will use this dilution factor (5) to automatically calculate the concentration of the sample and report the result.

If the operator does not enter the manual dilution factor, the result must be manually multiplied by the appropriate manual dilution factor (5) before reporting the result. If a diluted sample result is less than 0.1 mg/dL (1.7 µmol/L), do not report the result. Rerun using an appropriate dilution.

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

Calibration

For instructions on performing a calibration, refer to the ARCHITECT System Operations Manual, Section 6.

Calibration is stable for approximately 30 days (720 hours), with a blank adjustment after 15 days (360 hours). Calibration is required with each change in reagent lot. Verify calibration with at least 2 levels of controls according to the established quality control requirements for your laboratory. If control results fall outside acceptable ranges, recalibration may be necessary.

This assay may require recalibration after maintenance to critical parts or subsystems or after service procedures have been performed.

Quality Control Procedures

As appropriate, refer to your laboratory standard operating procedure(s) and/or quality assurance plan for additional quality control requirements and potential corrective actions.

- At least 2 levels of controls (low and high) are to be run every 24 hours.
- If more frequent control monitoring is required, follow the established quality control procedures for your laboratory.
- If quality control results do not meet the acceptance criteria defined by your laboratory, sample results may be suspect. Follow the established quality control procedures for your laboratory. Recalibration may be necessary. For troubleshooting information, refer to the ARCHITECT System Operations Manual, Section 10.
- Review quality control results and acceptance criteria following a change of reagent or calibrator lot.

Controls should be used according to the guidelines and recommendations of the control manufacturer. 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.

Quality Control Guidance

Refer to “Basic QC Practices” by James O. Westgard, Ph.D. for guidance on laboratory quality control practices.[13]

RESULTS

Calculation

The Total Bilirubin2 assay utilizes the Linear data reduction method to generate a calibration and results.

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.

Reportable Interval

Based on representative data for the limit of quantitation (LoQ) and the limit of detection (LoD), the ranges over which results can be reported are provided below according to the definitions from CLSI EP34, 1st ed.[14]

	mg/dL	µmol/L
Analytical Measuring Interval (AMI)[a]	0.1 - 25.0	1.7 - 427.5[d]
Extended Measuring Interval (EMI)[b]	25.0 - 125.0	427.5[d] - 2137.5[d]
Reportable Interval[c]	0.1 - 125.0	1.7 - 2137.5[d]

[a] AMI: The AMI is determined by the range of values in mg/dL (µmol/L) that demonstrated acceptable performance for linearity, imprecision, and bias. NOTE: The observed LoQ has been rounded up to the number of decimal places defined in the assay file.

[b] The EMI extends from the upper limit of quantitation (ULoQ) to the ULoQ × sample dilution.

[c] The reportable interval extends from the LoD (rounded up to the number of decimal places defined in the assay file) to the upper limit of the EMI.

[d] Value determined based on the dilution factor and instrument rounding.

NOTE: The Low Linearity value of the assay file corresponds to the lower limit of the AMI. Samples with total bilirubin value below 0.1 mg/dL (1.7 µmol/L) are reported as “<0.1 mg/dL” (“<1.7 µmol/L”).

LIMITATIONS OF THE PROCEDURE

- Results should be used in conjunction with other data; e.g., symptoms, results of other tests, and clinical impressions.
- Specimens with indican levels greater than 1 mg/dL may cause falsely elevated results with the Total Bilirubin2 assay. Refer to the SPECIFIC PERFORMANCE CHARACTERISTICS, Analytical Specificity, Interference section of this package insert for further information.
- Specimens with indocyanine green levels greater than 5 mg/L may cause falsely elevated results with the Total Bilirubin2 assay. Refer to the SPECIFIC PERFORMANCE CHARACTERISTICS, Analytical Specificity, Interference section of this package insert for further information. For patients undergoing evaluations involving the administration of indocyanine green, it is recommended that samples are drawn after indocyanine green has been eliminated.[15, 16]
- In very rare cases paraproteins may cause unreliable results.[17]
- In samples where the concentration of bilirubin is low or where conjugated bilirubin is the predominant form, the direct bilirubin assay may report results that are greater than results obtained using the total bilirubin assay. Under these circumstances, report the total bilirubin result for both the total and direct bilirubin assays.
- Substances that demonstrated interference with the Total Bilirubin2 assay are listed in the SPECIFIC PERFORMANCE CHARACTERISTICS, Analytical Specificity, Interference section of this package insert.
- Potential interference has not been evaluated for substances other than those described in the SPECIFIC PERFORMANCE CHARACTERISTICS, Analytical Specificity, Interference section of this package insert.
- SmartWashes for assays impacted by Total Bilirubin2 must be configured to avoid interference due to reagent carryover. See the INSTRUMENT PROCEDURE section of this package insert for the required assay file updates.

EXPECTED VALUES

It is recommended that each laboratory determine its own reference range based upon its particular locale and population characteristics.

Reference Range[18]

	Range (mg/dL)	Range (µmol/L)[a]
Premature		
0 to 1 day	< 8.0	< 136.8
1 to 2 days	< 12.0	< 205.2
3 to 5 days	< 16.0	< 273.6
Full-term		
0 to 1 day	2.0–6.0	34.2–102.6
1 to 2 days	6.0–10.0	102.6–171.0
3 to 5 days	1.5–12.0	25.7–205.2
Adults		
	0.3–1.2	5.1–20.5

[a] Alternate result units were calculated by Abbott and are not included in the citation provided.

SPECIFIC PERFORMANCE CHARACTERISTICS

Representative performance data are provided in this section. Results obtained in individual laboratories may vary.

Precision

Within-Laboratory Precision

A study was performed based on guidance from CLSI EP05-A3.[19] Testing was conducted using 3 lots of the Total Bilirubin2 reagents, 3 lots of the Consolidated Chemistry Calibrator, 1 lot of commercially available controls, and 3 instruments. Two controls and 3 human serum panels were tested in a minimum of 2 replicates, twice per day on 20 days on 3 reagent lot/calibrator lot/instrument combinations, where a unique reagent lot and a unique calibrator lot are paired with 1 instrument. The performance from a representative combination is shown in the following table.

			Within-Run (Repeatability)		Within-Laboratory[a]	
Sample	n	Mean (mg/dL)	SD	%CV	SD (Range[b])	%CV (Range[b])
Control Level 1	80	1.1	0.02	1.9	0.04 (0.02–0.04)	3.4 (1.8–3.4)
Control Level 2	80	4.2	0.04	0.9	0.09 (0.09–0.10)	2.1 (2.0–2.2)
Panel A	80	0.3	0.00	0.0	0.00 (0.00–0.03)	0.0 (0.0–9.2)
Panel B	80	13.3	0.09	0.7	0.11 (0.09–0.12)	0.8 (0.7–0.9)
Panel C	80	22.3	0.15	0.7	0.16 (0.16–0.18)	0.7 (0.7–0.8)

[a] Includes within-run, between-run, and between-day variability.

[b] Minimum and maximum SD or %CV across the 3 reagent lot/calibrator lot/instrument combinations.

			Within-Run (Repeatability)		Within-Laboratory[a]	
Sample	n	Mean (µmol/L)	SD	%CV	SD (Range[b])	%CV (Range[b])
Control Level 1	80	18.4	0.18	1.0	0.47 (0.43–0.47)	2.6 (2.3–2.6)
Control Level 2	80	72.2	0.37	0.5	1.41 (1.41–1.60)	1.9 (1.9–2.1)
Panel A	80	5.5	0.09	1.7	0.15 (0.15–0.18)	2.6 (2.6–3.2)
Panel B	80	226.9	1.47	0.6	1.76 (1.44–1.81)	0.8 (0.6–0.8)
Panel C	80	380.8	2.41	0.6	2.63 (2.63–3.03)	0.7 (0.7–0.8)

[a] Includes within-run, between-run, and between-day variability.

[b] Minimum and maximum SD or %CV across the 3 reagent lot/calibrator lot/instrument combinations.

Reproducibility

A study was performed based on guidance from CLSI EP05-A3.[19] Testing was conducted using 1 lot of the Total Bilirubin2 reagents, 1 lot of the Consolidated Chemistry Calibrator, 1 lot of commercially available controls, and 3 instruments. Each instrument was operated by a different technician. Two controls and 2 human serum panels were tested in a minimum of 3 replicates at 2 separate times per day on 5 different days*.

			Repeatability		Within-Laboratory[a]		Reproducibility[b]	
Sample	n	Mean (mg/dL)	SD	%CV	SD	%CV	SD	%CV
Control Level 1	84	1.1	0.02	1.6	0.02	2.1	0.02	2.2
Control Level 2	84	4.5	0.03	0.6	0.06	1.5	0.16	3.5
Panel B	84	13.4	0.08	0.6	0.10	0.7	0.57	4.3
Panel C	84	22.4	0.13	0.6	0.17	0.7	1.12	5.0

[a] Includes repeatability (within-run), between-run, and between-day variability.

[b] Includes repeatability (within-run), between-run, between-day, and between-instrument variability.

* One instrument includes results from 4 days of testing only.

Sample	n	Mean (µmol/L)	Repeatability SD	Repeatability %CV	Within-Laboratory[a] SD	Within-Laboratory[a] %CV	Reproducibility[b] SD	Reproducibility[b] %CV
Control Level 1	84	19.2	0.12	0.6	0.21	1.1	0.34	1.8
Control Level 2	84	76.0	0.47	0.6	1.17	1.5	2.80	3.7
Panel B	84	229.4	1.27	0.6	1.55	0.7	9.69	4.2
Panel C	84	383.4	2.20	0.6	2.85	0.7	19.06	5.0

[a] Includes repeatability (within-run), between-run, and between-day variability.

[b] Includes repeatability (within-run), between-run, between-day, and between-instrument variability.

[*] One instrument includes results from 4 days of testing only.

Accuracy

A study was performed to estimate the bias of the Total Bilirubin2 assay relative to material standardized to the Doumas Total Bilirubin reference method. Testing was conducted using 2 concentrations of the material across 3 lots of the Total Bilirubin2 reagents, 2 lots of the Consolidated Chemistry Calibrator, and 1 instrument. The bias ranged from -0.1% to 3.7%.

Lower Limits of Measurement

A study was performed based on guidance from CLSI EP17-A2.[20] Testing was conducted using 3 lots of the Total Bilirubin2 reagents on each of 2 instruments over a minimum of 3 days. The maximum observed limit of blank (LoB), limit of detection (LoD), and limit of quantitation (LoQ) values are summarized below.

	mg/dL	µmol/L
LoB[a]	0.02	0.3
LoD[b]	0.04	0.7
LoQ[c]	0.07	1.2

[a] The LoB represents the 95th percentile from n ≥ 60 replicates of zero-analyte samples.

[b] The LoD represents the lowest concentration at which the analyte can be detected with 95% probability based on n ≥ 60 replicates of low-analyte level samples.

[c] The LoQ is defined as the lowest concentration at which a maximum allowable precision of 20 %CV was met and was determined from n ≥ 60 replicates of low-analyte level samples.

Linearity

A study was performed based on guidance from CLSI EP06-A.[21]

This assay is linear across the analytical measuring interval of 0.1 to 25.0 mg/dL (1.7 to 427.5 µmol/L).

Analytical Specificity

Interference

Potentially Interfering Endogenous Substances

A study was performed based on guidance from CLSI EP07, 3rd ed.[22] Each substance was tested at 2 levels of the analyte (approximately 2 mg/dL and 15 mg/dL).

No significant interference (interference within ± 10%) was observed at the following concentrations.

No Significant Interference (Interference within ± 10%)		
	Interferent Level	
Potentially Interfering Substance	Default Units	Alternate Units
Hemoglobin	1000 mg/dL	10.0 g/L
Indican	1 mg/dL	33.9 µmol/L
Total protein	15 g/dL	150 g/L
Triglycerides	1500 mg/dL	16.9 mmol/L

Interference beyond ± 10% (based on 95% Confidence Interval [CI]) was observed at the concentration shown below for the following substance.

Interference beyond ± 10% (based on 95% Confidence Interval [CI])					
	Interferent Level		Analyte Level		
Potentially Interfering Substance	Default Units	Alternate Units	Default Units	Alternate Units	% Interference (95% CI)
Indican	2 mg/dL	67.7 µmol/L	2 mg/dL	34.2 µmol/L	17% (15%, 19%)

Potentially Interfering Exogenous Substances

A study was performed based on guidance from CLSI EP07, 3rd ed.[22] Each substance was tested at 2 levels of the analyte (approximately 2 mg/dL and 15 mg/dL).

No significant interference (interference within ± 10%) was observed at the following concentrations.

No Significant Interference (Interference within ± 10%)		
	Interferent Level	
Potentially Interfering Substance	Default Units	Alternate Units
4-Hydroxypropranolol glucuronide	0.2 mg/dL	4.43 µmol/L
Acetaminophen	160 mg/L	1059 µmol/L
Acetylcysteine	150 mg/L	920 µmol/L
Acetylsalicylic acid	30 mg/L	167 µmol/L
Ampicillin-Na	80 mg/L	215 µmol/L
Ascorbic acid	60 mg/L	341 µmol/L
Biotin	4250 ng/mL	17.4 µmol/L
Ca-dobesilate	60 mg/L	143 µmol/L
Cefoxitin	6600 mg/L	15.4 mmol/L
Cyanokit (hydroxocobalamin)	2259 mg/L	1678 µmol/L
Cyclosporine	2 mg/L	1.66 µmol/L
Doxycycline	20 mg/L	45.0 µmol/L
Eltrombopag	300 mg/L	678 µmol/L
Ibuprofen	220 mg/L	1067 µmol/L
Indocyanine green	5 mg/L	6.45 µmol/L
Iron dextran	60 mg/L	390 µmol/L
Levodopa	8 mg/L	40.6 µmol/L
Methyldopa	25 mg/L	118 µmol/L
Metronidazole	130 mg/L	759 µmol/L
Oxytetracycline	12 mg/L	26.0 µmol/L
Phenylbutazone	330 mg/L	1069 µmol/L
Propranolol	0.1 mg/dL	3.86 µmol/L
Rifampicin	50 mg/L	61.0 µmol/L
Sodium heparin	4 U/mL	N/A
Theophylline (1,3-dimethylxanthine)	60 mg/L	333 µmol/L

N/A = Not Applicable

Interference beyond ± 10% (based on 95% Confidence Interval [CI]) was observed at the concentrations shown below for the following substance.

Interference beyond ± 10% (based on 95% Confidence Interval [CI])					
	Interferent Level		Analyte Level		
Potentially Interfering Substance	Default Units	Alternate Units	Default Units	Alternate Units	% Interference (95% CI)
Indocyanine green	10 mg/L	12.9 µmol/L	2 mg/dL	34.2 µmol/L	9% (8%, 11%)

Interferences from medication or endogenous substances may affect results.[23]

Method Comparison

A study was performed based on guidance from CLSI EP09c, 3rd ed[24] using the Passing-Bablok regression method.

Total Bilirubin2 vs. Total Bilirubin on the ARCHITECT c System						
	n	Units	Correlation Coefficient	Intercept	Slope	Concentration Range
Serum	167	mg/dL	1.00	0.0	1.03	0.1–22.5
		(µmol/L)		(-1.1)	(1.03)	(1.4–384.5)
Neonatal serum	163	mg/dL	1.00	0.0	1.00	0.2–22.8
		(µmol/L)		(0.2)	(1.03)	(3.0–390.4)

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Key to Symbols

ISO 15223 Symbols	
	Consult instructions for use
	Manufacturer
	Sufficient for
	Temperature limitation
	Use by/Expiration date
IVD	*In Vitro* Diagnostic Medical Device
LOT	Lot Number
REF	List Number
SN	Serial number

Other Symbols	
DISTRIBUTED IN THE USA BY	Distributed in the USA by
FOR USE WITH	Identifies products to be used together
INFORMATION FOR USA ONLY	Information needed for United States of America only
PRODUCT OF IRELAND	Product of Ireland
R1	Reagent 1
R2	Reagent 2
Rx ONLY	For use by or on the order of a physician only (applicable to USA classification only).

Note for number formatting:

- A space is used as thousands separator (example: 10 000 specimens).
- A period is used to separate the integer part from the fractional part of a number written in decimal form (example: 3.12%).

The ARCHITECT c System family of instruments consists of c4000, c8000, and c16000 instruments.

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A summary of safety and performance (SSP) for this device is available at https://ec.europa.eu/tools/eudamed. This is the SSP location after the launch of European Database on Medical Devices. Search for the device using the UDI-DI provided on the outer packaging of the device.

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