ARCHITECT B12

REF 7K61-25
REF 7K61-35

en
B12
7K61
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Read Highlighted Changes: Revised November 2015.

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.

NAME

ARCHITECT B12

INTENDED USE

The ARCHITECT B12 assay is a Chemiluminescent Microparticle Intrinsic Factor assay for the quantitative determination of vitamin B12 in human serum and plasma on the ARCHITECT iSystem.

SUMMARY AND EXPLANATION OF THE TEST

Vitamin B12 (B12), a member of the corrin family, is a cofactor for the conversion of methylmalonyl Coenzyme-A (CoA) to succinoyl CoA. In addition, B12 is a cofactor in the synthesis of methionine from homocysteine, is implicated in the formation of myelin, and, along with folate, is required for DNA synthesis.[1, 2]

B12 is absorbed from food after binding to a protein called intrinsic factor which is produced by the stomach. Causes of vitamin B12 deficiency can be divided into three classes: nutritional deficiency, malabsorption syndromes, and other gastrointestinal causes. B12 deficiency can cause megaloblastic anemia (MA), nerve damage and degeneration of the spinal cord. Lack of B12, even mild deficiencies, damages the myelin sheath that surrounds and protects nerves, which may lead to peripheral neuropathy. The nerve damage caused by a lack of B12 may become permanently debilitating, if the underlying condition is not treated. People with intrinsic factor defects who do not get treatment eventually develop a MA called pernicious anemia (PA).[2]

The relationship between B12 levels and MA is not always clear in that some patients with MA will have normal B12 levels; conversely, many individuals with B12 deficiency are not afflicted with MA. Despite these complications, however, in the presence of MA (e.g., elevated mean corpuscular volume (MCV)) there is usually serum B12 or folate deficiency.[2, 3]

The true prevalence of B12 deficiency in the general population is unknown but increases with age. In one study,[4] fifteen percent of adults older than 65 years old had laboratory evidence of vitamin B12 deficiency.

A serum B12 level below the normal expected range may indicate that tissue B12 levels are becoming depleted. However, a B12 level in the low normal range does not ensure that B12 levels are healthy and symptomatic patients should be further evaluated with tests for holotranscobalamin,[5] homocysteine and methylmalonic acid.[6, 7]

There are a number of conditions that are associated with low serum B12 levels, including iron deficiency, normal near-term pregnancy, vegetarianism, partial gastrectomy/ileal damage, celiac disease, use of oral contraception, parasitic competition, pancreatic deficiency, treated epilepsy, and advancing age.[2, 8-11] Disorders associated with elevated serum B12 levels include renal failure, liver disease, and myeloproliferative diseases.[8, 12]

BIOLOGICAL PRINCIPLES OF THE PROCEDURE

The ARCHITECT B12 assay is a two-step assay with an automated sample pretreatment, for determining the presence of B12 in human serum and plasma using CMIA technology with flexible assay protocols, referred to as Chemiflex.

1. Sample and Pre-Treatment Reagent 1, Pre-Treatment Reagent 2, and Pre-Treatment Reagent 3 are combined.
2. An aliquot of the pre-treated sample is aspirated and transferred into a new Reaction Vessel (RV). The pre-treated sample, assay diluent, and intrinsic factor coated paramagnetic microparticles are combined. The B12 present in the sample binds to the intrinsic factor coated microparticles.
3. After washing, B12 acridinium-labeled conjugate is added to create a reaction mixture.
4. Following another wash cycle, Pre-Trigger and Trigger Solutions are added to the reaction mixture.
5. The resulting chemiluminescent reaction is measured as relative light units (RLUs). There is an inverse relationship between the amount of B12 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 B12 7K61

NOTE: Some kit sizes are not available in all countries or for use on all ARCHITECT iSystems. Please contact your local distributor.

REF	7K61-25	7K61-35
Σ	100	500
MICROPARTICLES	1 x 6.6 mL	1 x 27.0 mL
CONJUGATE	1 x 5.9 mL	1 x 26.3 mL
ASSAY DILUENT	1 x 10.0 mL	1 x 51.0 mL
PRE-TREATMENT REAGENT 1	1 x 27.0 mL	1 x 50.4 mL
PRE-TREATMENT REAGENT 2	1 x 3.2 mL	1 x 13.4 mL
PRE-TREATMENT REAGENT 3	1 x 3.3 mL	1 x 13.8 mL

MICROPARTICLES Intrinsic Factor (porcine) coated Microparticles in borate buffer with protein (bovine) stabilizers. Minimum Concentration: 0.1% solids. Preservative: antimicrobial agents.

CONJUGATE B12 acridinium-labeled Conjugate in MES buffer. Minimum concentration: 0.7 ng/mL. Preservative: ProClin.

ASSAY DILUENT B12 Assay Diluent containing borate buffer with EDTA. Preservative: antimicrobial agents.

PRE-TREATMENT REAGENT 1 B12 Pre-Treatment Reagent 1 containing 1.0 N sodium hydroxide with 0.005% potassium cyanide.

PRE-TREATMENT REAGENT 2 B12 Pre-Treatment Reagent 2 containing alpha monothioglycerol and EDTA.

PRE-TREATMENT REAGENT 3 B12 Pre-Treatment Reagent 3 containing cobinamide dicyanide in borate buffer with protein (avian) stabilizers. Preservative: sodium azide.

Other Reagents

MULTI-ASSAY MANUAL DILUENT 1 x 100 mL ARCHITECT Multi-Assay Manual Diluent, **REF** 7D82-50, containing phosphate buffered saline solution. Preservative: antimicrobial agent.

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.[13-16]

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

DANGER:	Contains sodium borate.
H360	May damage fertility or the unborn child.
Prevention	
P201	Obtain special instructions before use.
P280	Wear protective gloves / protective clothing / eye protection.
Response	
P308+P313	IF exposed or concerned: Get medical advice / attention.
Disposal	
P501	Dispose of contents / container in accordance with local regulations.

The following warnings and precautions apply to: **PRE-TREATMENT REAGENT 3**

DANGER:	Contains sodium borate and sodium azide.
H360	May damage fertility or the unborn child.
EUH032	Contact with acids liberates very toxic gas.
Prevention	
P201	Obtain special instructions before use.
P280	Wear protective gloves / protective clothing / eye protection.
Response	
P308+P313	IF exposed or concerned: Get medical advice / attention.
Disposal	
P501	Dispose of contents / container in accordance with local regulations.

The following warnings and precautions apply to: **PRE-TREATMENT REAGENT 2**

WARNING:	Contains monothioglycerol.
H315	Causes skin irritation.
H319	Causes serious eye irritation.
Prevention	
P264	Wash hands thoroughly after handling.
P280	Wear protective gloves / protective clothing / eye protection.
Response	
P305+P351+P338	IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P337+P313	If eye irritation persists: Get medical advice / attention.
P302+P352	IF ON SKIN: Wash with plenty of water.
P332+P313	If skin irritation occurs: Get medical advice / attention.
P362+P364	Take off contaminated clothing and wash it before reuse.

The following warnings and precautions apply to: **PRE-TREATMENT REAGENT 1**

DANGER:	Contains sodium hydroxide.
H314	Causes severe skin burns and eye damage.
H290	May be corrosive to metals.
Prevention	
P234	Keep only in original container.
P260	Do not breathe mist / vapors / spray.
P264	Wash hands thoroughly after handling.
P280	Wear protective gloves / protective clothing / eye protection.
Response	
P301+P330+P331	IF SWALLOWED: Rinse mouth. Do NOT induce vomiting.
P305+P351+P338	IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P303+P361+P353	IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water / shower.
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.

The following warnings and precautions apply to: CONJUGATE	
	
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.

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.**
  - Prolonged exposure of B12 Pre-Treatment Reagent 1 to air may compromise performance.
- 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 B12 assay file must be installed on the ARCHITECT iSystem from an ARCHITECT iSystem Assay CD-ROM 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
pg/mL	0.7378	pmol/L

SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS

Specimen Types

Verified specimen types to be used with this assay:

Glass	Plastic
Serum	Serum
	Serum separator tubes
	Lithium heparin plasma separator
	Sodium heparin
	Dipotassium EDTA

- Other specimen collection tube types have not been tested with this assay.
- Performance has not been established for 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
  - pooled
  - hemolyzed
  - 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, 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.
- 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	≤ 3 days
	2-8°C	≤ 7 days

Specimens may be stored on or off the clot, red blood cells, or separator gel.

If testing will be delayed more than 3 days for specimens stored at room temperature or more than 7 days for specimens stored at 2-8°C, remove serum or plasma from the clot, red blood cells, or separator gel and store at -20°C or colder.

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

7K61 ARCHITECT B12 Reagent Kit

Materials Required but not Provided

- ARCHITECT B12 Assay file obtained from the ARCHITECT iSystem e-Assay CD-ROM found on www.abbottdiagnostics.com.
- 7K61-01 ARCHITECT B12 Calibrators
- 7K61-10 ARCHITECT B12 Controls
- 7D82-50 ARCHITECT Multi-Assay Manual Diluent
- 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.
  - 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, calibrators and controls and for general operating procedures, refer to the ARCHITECT System Operations Manual, Section 5.
- Minimum sample cup volume is calculated by the system and printed on the Orderlist report. To minimize the effects of evaporation, verify 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 first test: 87 µL

    Sample volume for each additional test from same sample cup: 37 µL
  - ≤ 3 hours on board:

    Sample volume for first test: 150 µL

    Sample volume for each additional test from same sample cup: 37 µL
  - > 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 ARCHITECT B12 Calibrators and Controls.
  - Mix calibrator(s) and controls by gentle inversion before use.
  - Hold bottles **vertically** and dispense recommended volumes into each respective sample cup.
  - Recommended volumes:

    for each calibrator: 3 drops

    for each control: 3 drops

- Load samples.
  - For information on loading samples, refer to the ARCHITECT System Operations Manual, Section 5.
- Press RUN.
- 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 B12 value exceeding 2000 pg/mL (1476 pmol/L) are flagged with the code ">2000" when working in pg/mL (">1476" when working in pmol/L) and may be diluted using either the Automated Dilution Protocol or the Manual Dilution Procedure.

Automated Dilution Protocol

The system performs a 1:3 dilution of the specimen and automatically calculates the concentration of the specimen before dilution and reports the result.

Manual Dilution Procedure

Suggested dilution: 1:4

Suggested dilution for specimens that generate repeated (2 or more) "3350 Unable to process test-aspiration error for (Sample Pipettor) at (RV 24)" errors: 1:2

1. For a 1:4 dilution, add 100 µL of the patient specimen to 300 µL of ARCHITECT Multi-Assay Manual Diluent.
2. For a 1:2 dilution, add 100 µL of the patient specimen to 100 µL of ARCHITECT Multi-Assay Manual Diluent.
3. 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 > 83 pg/mL (61 pmol/L) 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: 0-2000 pg/mL (0-1476 pmol/L).
- Once an ARCHITECT B12 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 B12 assay is that a single replicate 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.

Control values must be within the ranges specified in the control package insert. If a control result is out of its specified range, any test results generated since the last acceptable control results must be evaluated to determine if sample results may have been adversely affected. Adversely affected test results are invalid, and these samples must be retested. For troubleshooting information, refer to the ARCHITECT System Operations Manual, Section 10.

Verification of Assay Claims

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

The ARCHITECT B12 assay belongs to method group 1.

RESULTS

Calculation

The ARCHITECT B12 assay utilizes a 4 Parameter Logistic Curve fit data reduction method (4PLC, Y-weighted) to generate a calibration curve.

For information on alternate result units, refer to the **INSTRUMENT PROCEDURE, Alternate Result Units** section of this package insert.

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.

LIMITATIONS OF THE PROCEDURE

- Results should be used in conjunction with other data; e.g., symptoms, results of other tests, and clinical impressions.
- The diagnosis of B12 deficiency cannot be solely based on serum or plasma B12 levels. Further testing for folic acid, intrinsic factor blocking antibodies, holotranscobalamin,[5] homocysteine, and/or methylmalonic acid is suggested for symptomatic patients with hematological or neurological abnormalities.[6, 7]
- If the B12 results are inconsistent with clinical evidence, additional testing is recommended to confirm the result.
- Hemolysis has been demonstrated to exhibit negative interference in this B12 assay. Hemolyzed specimens should not be analyzed.
- Specimens containing above normal protein concentrations may generate repeated (2 or more) "3350 Unable to process test-aspiration error for (Sample Pipettor) at (RV 24)" errors and should be quantified using the Automated Dilution Protocol or Manual Dilution Procedure (1:2).
- Heterophilic antibodies and rheumatoid factor (RF) in human serum can react with reagent immunoglobulins, interfering with *in vitro* immunoassays. Patients routinely exposed to animals or to animal serum products can be prone to this interference and anomalous values may be observed. Additional information may be required for diagnosis.[17]
- The assay is designed to test human serum and plasma. Specimens tested in other matrices may not give accurate results.
- Refer to the **SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS** section of this package insert for specimen limitations.

EXPECTED VALUES

B12 Normals

It is recommended that each laboratory establish its own range, which may be unique to the population it serves depending upon geographical, patient, dietary, or environmental factors.

A study was performed based on guidance from Clinical and Laboratory Standards Institute (CLSI) document C28-A2.[18] Serum specimens from 143 individuals with normal mean corpuscular volume, homocysteine, and folate results were assayed for B12 using the ARCHITECT B12 assay. The B12 concentration range for this population was 141 to > 1218 pg/mL (104 to > 899 pmol/L) with a mean of 407 pg/mL (300 pmol/L). The central 95% of the sample population is defined below:

Expected Range	187-883 pg/mL	(138-652 pmol/L)

B12 Indeterminates

Levels above 300 or 400 pg/mL (221 or 295 pmol/L) are rarely associated with B12 deficiency induced hematological or neurological disease, respectively. Further testing is suggested for symptomatic patients with B12 levels between 100 and 300 pg/mL (74 and 221 pmol/L) (hematological abnormalities), and between 100 and 400 pg/mL (74 and 295 pmol/L) (neurological abnormalities).[6, 7]

SPECIFIC PERFORMANCE CHARACTERISTICS

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

Precision

The ARCHITECT B12 assay is designed to have a Total CV of ≤ 11% for concentrations in the range of the low, medium, and high controls. A 20-day precision study was performed for the ARCHITECT B12 assay based on guidance from the CLSI document EP5-A2.[19] Testing was conducted at Abbott Laboratories using three ARCHITECT B12 assay reagent lots, two calibrator lots, one control lot, and two instruments. Four levels of controls and panels were assayed in replicates of three at two separate times of day for 20 different days. The data are summarized in the following table.

				Within Run		Within Laboratory Precision (Total)	
Instrument	Sample	n	Mean (pg/mL)	SD	%CV	SD	%CV
1	Serum Panel	360	262	12.6	4.8	16.3	6.2
	Low Control	354	246	13.8	5.6	16.7	6.8
	Medium Control	355	424	14.3	3.4	16.8	4.0
	High Control	359	890	36.0	4.0	38.9	4.4
2	Serum Panel	357	248	11.6	4.7	13.3	5.4
	Low Control	356	241	10.4	4.3	12.9	5.4
	Medium Control	352	408	13.3	3.3	15.5	3.8
	High Control	355	885	23.9	2.7	29.7	3.4

Accuracy by WHO

A study was conducted to evaluate the accuracy of the ARCHITECT B12 assay using the B12 World Health Organization International Standard 03/178. The assay demonstrated a -3.6% difference from the target value of 480 pg/mL (354 pmol/L).

Sensitivity

Sensitivity is defined as the Limit of Blank (LoB), Limit of Detection (LoD), and Limit of Quantitation (LoQ) for the ARCHITECT B12 assay. The assay is designed to have an LoQ of ≤ 150 pg/mL (≤ 111 pmol/L). A study conducted based on guidance from CLSI document EP17-A[20] produced an LoB of 83 pg/mL (61 pmol/L), an LoD of 125 pg/mL (92 pmol/L) and LoQ of 125 pg/ mL (92 pmol/L).

Specificity

The ARCHITECT B12 assay is designed to have an interference (difference) less than the LoD of the assay with cobinamide, a B12 analogue. The specificity of the ARCHITECT B12 assay was determined by studying the cross reactivity with cobinamide. A human serum specimen at approximately 230 pg/mL (168 pmol/L) was supplemented with cobinamide at 9000 pg/mL and the resulting interference was 4 pg/mL (3 pmol/L).

Interference

At the concentrations listed below, bilirubin (conjugated and unconjugated), total protein, and triglycerides showed less than 10% interference in the ARCHITECT B12 assay for low samples (concentration range: 150 pg/mL to 250 pg/mL (111 pmol/L to 184 pmol/L)) and higher samples (concentration range: > 500 pg/mL (> 369 pmol/L)):

Bilirubin	< 25.1 mg/dL
Total Protein	< 12 g/dL
Triglycerides	< 3325 mg/dL

Hemolyzed specimens should not be analyzed; refer to the **LIMITATIONS OF THE PROCEDURE** section of this package insert.

Accuracy by Correlation

A study was conducted based on guidance from CLSI document EP9-A2.[21] Three hundred and twenty nine serum specimens were tested for the determination of B12 using the ARCHITECT B12 assay and a commercially available diagnostic kit. The specimen testings are shown in the following table*.

Abbott ARCHITECT B12 vs AxSYM B12

Method	Number of Specimens	Intercept	Slope	Correlation Coefficient
Least Squares Linear Regression	329	-2.05	1.01	0.99
Passing-Bablok Linear Regression[22]	329	21.96	0.95	0.99

In this evaluation, serum specimens tested ranged from 113 to 2769 pg/mL (83 to 2043 pmol/L) by the ARCHITECT B12 assay, and from 93.5 to 2655.8 pg/mL (69.1 to 1959.5 pmol/L) by the comparator assay.

* Representative data, results in individual labs may vary from these data.

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Key to Symbols

	Consult instructions for use
	Manufacturer
	Sufficient for
	Temperature limitation
	Use by/Expiration date
ASSAY DILUENT	Assay Diluent
CONJUGATE	Conjugate
CONTAINS: AZIDE	Contains Sodium Azide. Contact with acids liberates very toxic gas.
CONTROL NO.	Control Number
DANGER: REPRODUCTIVE HAZARD	Danger: Reproductive Hazard
IVD	*In Vitro* Diagnostic Medical Device
LOT	Lot Number
MICROPARTICLES	Microparticles
MULTI-ASSAY MANUAL DILUENT	Multi-Assay Manual Diluent
PRE-TRIGGER SOLUTION	Pre-Trigger Solution
PRE-TREATMENT REAGENT 1	Pre-Treatment Reagent 1
PRE-TREATMENT REAGENT 2	Pre-Treatment Reagent 2
PRE-TREATMENT REAGENT 3	Pre-Treatment Reagent 3
PRODUCT OF IRELAND	Product of Ireland
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
WARNING: SENSITIZER	Warning: May cause an allergic reaction.
WARNING: SEVERE IRRITANT	Warning: Severe Irritant
WASH BUFFER	Wash Buffer

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