Instructions for Use - Atellica IM FoI

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SIEMENS Healthineers

Folate (Fol)

Current Revision and Date[a]	Rev. 05, 2023-11	
Product Name	Atellica IM Folate (Fol)	REF 10995572 (140 tests) REF 10995573 (700 tests)
Abbreviated Product Name	Atellica IM Fol	
Test Name/ID	Fol FolSerum	
Systems	Atellica IM Analyzer	
Materials Required but Not Provided	Atellica IM Fol DTT/REL	REF 10995576
	Atellica IM APW1	REF 10995458
Optional Materials	Atellica IM RBC Fol	REF 10995440
	Atellica IM Fol DIL	REF 10995574
	Atellica IM Fol MCM	REF 10995575
Specimen Types	Serum, heparinized whole blood	
Sample Volume	100 µL	
Measuring Interval	0.35–24.00 ng/mL (0.79–54.36 nmol/L)	

[a] A vertical bar in the page margin indicates technical content that differs from the previous version.

CE 0197

Intended Use

The Atellica® IM Folate (Fol) assay is for *in vitro* diagnostic use in the quantitative determination of folate in human serum and red blood cells using the Atellica® IM Analyzer.

Summary and Explanation

Folates are compounds of pteroylglutamic acid (PGA) that function as coenzymes in metabolic reactions involving the transfer of single-carbon units from a donor to a recipient compound. Folate, with vitamin B_{12}, is essential for DNA synthesis, which is required for normal red blood cell maturation.[1] Humans obtain folate from dietary sources including fruits, green and leafy vegetables, yeast, and organ meats.[2] Folate is absorbed through the small intestine and stored in the liver. Measurements of folate are used as an aid in the diagnosis and treatment of folate deficiency.[3]

Low folate intake, malabsorption as a result of gastrointestinal diseases, pregnancy, and drugs such as phenytoin are causes of folate deficiency.[4] Folate deficiency is also associated with chronic alcoholism.[5] Folate and vitamin B_{12} deficiency impair DNA synthesis, causing macrocytic anemias. These anemias are characterized by abnormal maturation of red blood cell precursors in the bone marrow, the presence of megaloblasts, and decreased red blood cell survival.[1]

Since both folate and vitamin B_{12} deficiency can cause macrocytic anemia, appropriate treatment depends on the differential diagnosis of the deficiency. Serum folate measurement provides an early index of folate status.[2] However, folate is much more concentrated in red blood cells than in serum so the red blood cell folate measurement more closely reflects tissue stores.[5] Red blood cell folate concentration is considered the most reliable indicator of folate status.[2]

Principles of the Procedure

The Atellica IM Fol assay is a competitive immunoassay using direct chemiluminescent technology. Folate in the patient sample competes with acridinium-ester-labeled folate in the Lite Reagent for a limited amount of biotin-labeled folate binding protein. Biotin-labeled folate binding protein binds to avidin that is covalently coupled to paramagnetic particles in the Solid Phase. In the Atellica IM Fol assay, the sample is pretreated to release the folate from endogenous binding proteins in the sample.

An inverse relationship exists between the amount of folate present in the patient sample and the amount of relative light units (RLUs) detected by the system.

Reagents

Material Description	Storage	Stability[a]
Atellica IM Fol ReadyPack® primary reagent pack **Lite Reagent** 9.1 mL/reagent pack Folate labeled with acridinium ester (~9.8 ng/mL) in buffer; bovine serum albumin; sodium azide (0.1%); preservatives **Solid Phase** 18.2 mL/reagent pack Purified avidin (~20 µg/mL) covalently coupled to paramagnetic particles in buffer; human serum albumin; preservatives **Folate Binding Protein** 9.1 mL/reagent pack Purified folate binding protein (~1.0 µg/mL) covalently coupled to biotin in buffer; bovine serum albumin; preservatives	Unopened at 2–8°C Onboard	Until expiration date on product 14 days

Material Description	Storage	Stability[a]
Atellica IM Fol CAL 3.0 mL/vial; lyophilized After reconstitution, low or high levels of N-5-methyltetrahydrofolic acid; buffer; human serum albumin; sodium azide (< 0.1%); preservatives	Lyophilized at 2–8°C	Until expiration date on product
	Reconstituted at 2–8°C	7 days
	Reconstituted at room temperature	8 hours
	Reconstituted at ≤ -20°C	28 days
Atellica IM Fol DTT/Releasing Agent[b] **DTT** 8.0 mL/vial Dithiothreitol (~95 mg/mL in liquid form) **Releasing Agent** 4.0 mL/vial Sodium hydroxide (~1.1 N)	At 2–8°C	Until expiration date on product
	Onboard in an ancillary reagent pack[c]	108 hours
Atellica IM RBC Fol[d, e] **RBC Folate Ascorbic Acid** Lyophilized ascorbic acid (~0.30 g/vial) **RBC Folate Ascorbic Acid Diluent** 30.0 mL/vial Bovine serum albumin; buffer; sodium azide (< 0.1%); preservatives	Unopened at 2–8°C	Until expiration date on product
	Reconstituted at 2–8°C	30 days
Atellica IM Fol DIL ReadyPack ancillary reagent pack[e] 10.0 mL/reagent pack Human plasma; sodium azide (< 0.1%); preservatives	Unopened at 2–8°C	Until expiration date on product
	Onboard	28 days
Atellica IM APW1 ReadyPack ancillary reagent pack[c] 25.0 mL/pack 0.4 N sodium hydroxide	Unopened at 2–8°C	Until expiration date on product
	Onboard	14 days

[a] Refer to *Storage and Stability*.
[b] Refer to *Materials Required but Not Provided*.
[c] Refer to *Preparing the Reagents*.
[d] Refer to *Optional Materials*.
[e] Refer to *Preparing the Red Blood Cell Hemolysate* in the *Preparing the Samples* section.

Warnings and Precautions

For *in vitro* diagnostic use.

For Professional Use.

CAUTION

Federal (USA) law restricts this device to sale by or on the order of a licensed healthcare professional.

Safety data sheets (SDS) available on siemens-healthineers.com.

H290, H314
P234, P264, P280, P301+P330+P331, P303+P361+P353, P310, P305+P351+P338, P390, P501

Danger!
May be corrosive to metals. Causes severe skin burns and eye damage. Keep only in original container. Wash hands thoroughly after handling. Wear protective gloves/protective clothing/eye protection/face protection. IF SWALLOWED: rinse mouth. Do NOT induce vomiting. IF ON SKIN (or hair): Remove/Take off immediately all contaminated clothing. Rinse skin with water/shower. Immediately call a POISON CENTER or doctor/physician. IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Absorb spillage to prevent material damage. Dispose of contents and container in accordance with all local, regional, and national regulations.
Contains: sodium hydroxide (Atellica IM Fol Releasing Agent)

H290, H319, H315
P234, P264, P280, P337+P313, P390, P501

Warning!
May be corrosive to metals. Causes serious eye irritation. Causes skin irritation.
Keep only in original container. Wash hands thoroughly after handling. Wear protective gloves/protective clothing/eye protection/face protection. If eye irritation persists: Get medical advice/attention. Absorb spillage to prevent material damage. Dispose of contents and container in accordance with all local, regional, and national regulations.
Contains: sodium hydroxide (Atellica IM APW1)

H412
P273, P501

Harmful to aquatic life with long lasting effects.
Avoid release to the environment. Dispose of contents and container in accordance with all local, regional, and national regulations.
Contains: sodium azide (in Atellica IM Fol CAL)

H360F, H360D
P201, P280, P308+P313

Danger!
May damage fertility. May damage the unborn child.
Obtain special instructions before use. Wear protective gloves/protective clothing/eye protection/face protection. IF exposed or concerned: Get medical advice/attention.
Contains: disodium tetraborate decahydrate (Atellica IM FOL Solid phase and FOL Binding Protein)

Warning! Potential Biohazard

Contains human source material.

No known test method can ensure that products derived from human source materials will not transmit infection. These materials should be handled using good laboratory practices and universal precautions.[6-8]

CAUTION

This device contains material of animal origin and should be handled as a potential carrier and transmitter of disease.

Contains sodium azide as a preservative. Sodium azide can react with copper or lead plumbing to form explosive metal azides. On disposal, flush reagents with a large volume of water to prevent buildup of azides. Disposal into drain systems must be in compliance with prevailing regulatory requirements.

Dispose of hazardous or biologically contaminated materials according to the practices of your institution. Discard all materials in a safe and acceptable manner and in compliance with prevailing regulatory requirements.

Note For information about reagent preparation, refer to *Preparing the Reagents* in the *Procedure* section.

Note For information about calibrator preparation, refer to *Preparing the Calibrators*.

Storage and Stability

Store reagents in an upright position. Protect the product from heat and light sources. Unopened reagents are stable until the expiration date on the product when stored at 2–8°C.

Store calibrators in an upright position. Lyophilized calibrators are stable until the expiration date on the product when stored at 2–8°C. Reconstituted calibrators are stable for 7 days at 2–8°C or 8 hours at room temperature. Freeze reconstituted product at ≤ -20°C for up to 28 days.

Store Atellica IM Fol DTT/Releasing Agent in an upright position. Unopened Atellica IM Fol DTT/Releasing Agent is stable until the expiration date on the product when stored at 2–8°C.

Store Atellica IM RBC Fol in an upright position. Unopened Atellica IM RBC Fol is stable until the expiration date on the product when stored at 2–8°C. Reconstituted material is stable for 30 days at 2–8°C.

Store Atellica IM Fol DIL in an upright position. Unopened Atellica IM Fol DIL is stable until the expiration date on the product when stored at 2–8°C.

Store Atellica IM APW1 in an upright position. Unopened Atellica IM APW1 is stable until the expiration date on the product when stored at 2–8°C.

Do not use products beyond the expiration date printed on the product labeling.

Onboard Stability

Atellica IM Fol ReadyPack primary reagent packs are stable onboard the system for 14 days. Discard reagents at the end of the onboard stability interval.

Atellica IM Fol DTT/Releasing Agent is stable onboard the system for 108 hours.

Atellica IM Fol DIL is stable onboard the system for 28 days.

Atellica IM APW1 is stable onboard the system for 14 days.

Do not use products beyond the expiration date printed on the product labeling.

Specimen Collection and Handling

Serum and red blood cells are the recommended sample types for this assay.

Note Folates are light-sensitive. Minimize exposure to light during sample handling and storage.[10]

Collecting the Specimen

- Observe universal precautions when collecting specimens. Handle all specimens as if they are capable of transmitting disease.[8]
- Follow recommended procedures for collection of diagnostic blood specimens by venipuncture.[11]
- Follow the instructions provided with your specimen collection device for use and processing.[12]
- Allow blood specimens to clot completely before centrifugation.[9]
- Keep tubes capped at all times.[9]

Storing the Specimen

- Store serum samples at room temperature for no longer than 8 hours.
- Tightly cap and refrigerate specimens at 2–8°C if the assay is not completed within 8 hours.
- Freeze serum samples at ≤ -20°C if the assay is not completed within 48 hours.
- Freeze serum samples only 1 time and mix thoroughly after thawing. Frozen specimens can remain frozen up to 30 days. Do not store in a frost-free freezer. If serum samples will be stored for longer than 30 days, then they must be frozen at ≤ -80°C.
- If testing is not done within 24 hours for whole blood specimens, determine the hematocrit and freeze the whole blood specimen or hemolysate. Frozen whole blood specimens can be stored at -20°C for up to 2 months. Sample hemolysates prepared with the reconstituted RBC Folate Ascorbic Acid can be stored at -20°C for up to 3 months. Do not store whole blood specimens or hemolysates in a frost-free freezer. Freeze specimens only 1 time and mix thoroughly after thawing.

The handling and storage information provided here is based on data or references maintained by the manufacturer. It is the responsibility of the individual laboratory to use all available references and/or its own studies when establishing alternate stability criteria to meet specific needs.

Transporting the Specimen

Package and label specimens for shipment in compliance with applicable federal and international regulations covering the transport of clinical specimens and etiological agents.

Preparing the Samples

This assay requires 100 µL of sample for a single determination. This volume does not include the unusable volume in the sample container or the additional volume required when performing duplicates or other tests on the same sample. For information about determining the minimum required volume, refer to the online help.

The sample volume required to perform onboard dilution differs from the sample volume required to perform a single determination. Refer to *Dilutions*.

Note Do not use specimens with apparent contamination.

Before placing samples on the system, ensure that samples are free of:

- Bubbles or foam.
- Fibrin or other particulate matter.

Note Remove particulates by centrifugation according to CLSI guidance and the collection device manufacturer's recommendations.[9]

Note For a complete list of appropriate sample containers, refer to the online help.

Preparing the Red Blood Cell Hemolysate

Note Folates are light sensitive. Minimize exposure to light during sample handling and storage.[10]

1. Reconstitute the RBC Folate Ascorbic Acid by adding the contents of 1 vial of RBC Folate Ascorbic Acid Diluent to the lyophilized RBC Folate Ascorbic Acid.
2. Let the reconstituted mixture stand at room temperature for 15 minutes and mix by inverting the bottle occasionally.
3. Collect the sample in a tube containing heparin or EDTA.
4. Invert the sample several times to mix.

5. Determine and record the hematocrit.
6. Dispense 1.0 mL of reconstituted RBC Folate Ascorbic Acid into a test tube or sample cup.
7. Add 50 µL of the sample into the RBC Folate Ascorbic Acid.
8. Cap and invert the tube several times or vortex gently to mix. Avoid foaming.
9. Let the hemolysate stand protected from light, at room temperature, **for a minimum of 90 minutes, but less than 3 hours**.
10. **Do not mix the hemolysate again before placing the sample on the system.**

Note Do not dilute the RBC hemolysate. Freeze the hemolysate at ≤ -20°C immediately if testing cannot be completed within 4 hours from the time you finish preparing the hemolysate. Hemolysates can be stored at ≤ -20°C for up to 3 months. Do not store in a frost-free freezer.

If the hemolysate is frozen, thaw the hemolysate and mix it by inverting the tube several times. Let the hemolysate stand for 30 minutes at room temperature before testing. Test the hemolysate within 3 hours after thawing.

Procedure

Materials Provided

The following materials are provided:

REF	Contents	Number of Tests
10995572	1 ReadyPack primary reagent pack containing Atellica IM FoI Lite Reagent, Solid Phase, and Folate Binding Protein Atellica IM FoI (whole blood) master curve and test definition MC TDEF Atellica IM FoISR (serum) master curve and test definition MC TDEF 2 vials Atellica IM FoI CAL low calibrator CAL L 2 vials Atellica IM FoI CAL high calibrator CAL H Atellica IM FoI CAL calibrator lot-specific value sheet CAL LOT VAL	140
10995573	5 ReadyPack primary reagent packs containing Atellica IM FoI Lite Reagent, Solid Phase, and Folate Binding Protein Atellica IM FoI (whole blood) master curve and test definition MC TDEF Atellica IM FoISR (serum) master curve and test definition MC TDEF 2 vials Atellica IM FoI CAL low calibrator CAL L 2 vials Atellica IM FoI CAL high calibrator CAL H Atellica IM FoI CAL calibrator lot-specific value sheet CAL LOT VAL	700

Materials Required but Not Provided

The following materials are required to perform this assay, but are not provided:

REF	Description	
	Atellica IM Analyzer[a]	
10995576	Atellica IM Fol DTT/REL (releasing agent)	3 x 8.0 mL/vial DTT 3 x 4.0 mL/vial Releasing Agent REL 3 empty ReadyPack ancillary reagent packs
10995458	Atellica IM APW1 (probe wash)	2 ReadyPack ancillary reagent packs containing 25.0 mL/pack WASH

[a] Additional system fluids are required to operate the system: Atellica IM Wash, Atellica IM Acid, Atellica IM Base, and Atellica IM Cleaner. For system fluid instructions for use, refer to the Document Library.

Optional Materials

The following materials may be used to perform this assay, but are not provided:

REF	Description	
10995440	Atellica IM RBC Fol (ascorbic acid/diluent)	4 x 0.30 g/vial Ascorbic Acid ASCORBIC ACID 4 x 30.0 mL/vial Ascorbic Acid Diluent ASCORBIC ACID DIL
10995574	Atellica IM Fol DIL (diluent)	2 ReadyPack ancillary reagent packs containing 10.0 mL/pack DIL
10995575	Atellica IM Fol MCM (master curve material)	6 x 1.0 mL levels of master curve material MCM

Assay Procedure

The system automatically performs the following steps:

1. Dispenses 100 µL of sample into a cuvette.
2. Dispenses 35 µL of DTT/Releasing Agent, then incubates for 5 minutes at 37°C.
3. Dispenses 65 µL of Folate Binding Protein and 130 µL of Solid Phase, then incubates for 5 minutes at 37°C.
4. Dispenses 65 µL of Lite Reagent, then incubates for 3 minutes at 37°C.
5. Separates, aspirates, then washes the cuvettes with Atellica IM Wash.
6. Dispenses 300 µL each of Atellica IM Acid and Atellica IM Base to initiate the chemiluminescent reaction.
7. Reports results.

Preparing the Reagents

All reagents are liquid and ready to use, with the exception of the Atellica IM Fol DTT/Releasing Agent. Before loading primary reagent packs onto the system, mix them by hand and visually inspect the bottom of the reagent pack to ensure that all particles are resuspended. For information about preparing the reagents for use, refer to the online help.

Preparing the DTT/Releasing Agent

Note Careful preparation of DTT/Releasing Agent is required to obtain accurate and consistent results since the absolute amount of DTT delivered to each test can affect results. The prepared DTT/Releasing Agent must be used within 108 hours after preparation.

1. Carefully transfer the contents of 1 vial of Releasing Agent into 1 vial of DTT. For convenience, the Releasing Agent can be poured or transferred by pipette into the DTT vial.
2. Firmly screw the cap on the DTT vial and invert the vial several times to mix.
3. Pour or pipette the entire contents of the DTT vial into the disposable ancillary reagent pack provided.
4. Place a pack seal on the disposable ancillary reagent pack. Ensure that the seal is centered over the openings of the pack, and press firmly on the adhesive portion of the seal.

Note DTT/Releasing Agent ReadyPack ancillary reagent packs are lot-number-specific. Do not use packs from one lot of DTT/Releasing Agent with any other lot of DTT/Releasing Agent.

Preparing the System

Ensure that the system has sufficient reagent packs loaded in the reagent compartment. The system automatically mixes reagent packs to maintain homogeneous suspension of the reagents. For information about loading reagent packs, refer to the online help.

For automated dilutions, ensure that Atellica IM FoI DIL is loaded on the system.

Master Curve Definition

Before initiating calibration on each new lot of reagent, load the assay master curve and test definition values by scanning the MC TDEF 2D barcodes. For loading instructions, refer to the online help.

Performing Calibration

For calibration of the Atellica IM FoI assay, use the calibrators provided with each kit.

Do not pour the calibrators back into the vials after calibration because evaporation could occur, which may affect performance.

Do not refill calibrator sample cups when the contents are depleted. If required, dispense fresh calibrators.

Calibration Frequency

Perform a calibration if one or more of the following conditions exist:

- When changing lot numbers of primary reagent packs.
- At the end of the lot calibration interval, for a specified lot of calibrated reagent on the system.
- At the end of the pack calibration interval, for calibrated reagent packs on the system.
- When indicated by quality control results.
- After major maintenance or service, if indicated by quality control results.

At the end of the onboard stability interval, replace the reagent pack on the system with a new reagent pack. Recalibration is not required, unless the lot calibration interval is exceeded.

Stability Interval	Days
Lot Calibration	14
Pack Calibration	7
Reagent Onboard Stability	14

For information about lot calibration and pack calibration intervals, refer to the online help.

Follow government regulations or accreditation requirements for calibration frequency. Individual laboratory quality control programs and procedures may require more frequent calibration.

Preparing the Calibrators

Prepare calibrators using the following steps:

1. Add 3.0 mL of special reagent water into each vial using a precision pipet. Replace cap.

   Note For information about special reagent water requirements, refer to the online help.

2. Let the vials stand for 15–20 minutes at room temperature to allow the lyophilized material to dissolve.
3. Gently mix and invert the vials to ensure homogeneity of the material.
4. For extended storage, aliquot and seal tightly. Store reconstituted material according to stability limits specified in *Storing the Specimen*. Do not store in a frost-free freezer.

   Note Before using frozen calibrators, allow the material to completely thaw. Gently mix and invert the vials to ensure homogeneity of the material. Use immediately and discard any remaining material.

Note Use calibrators within the stability limits specified in *Storing the Specimen* and discard any remaining material.

Calibration Procedure

The required sample volume for testing depends on several factors. For information about sample volume requirements, refer to the online help.

Use the following lot-specific materials to perform calibration:

- For the master curve and assay test definitions, refer to the lot-specific master curve and test definition sheet MC TDEF provided with the assay reagents.
- Calibrators provided in an assay kit must only be used with reagents from that assay kit lot. Do not use calibrators from one assay kit with reagents from a different assay kit lot.
- For the calibrator definitions, refer to the lot-specific value sheet CAL LOT VAL provided with the calibrator materials.
- Generate lot-specific barcode labels to use with the calibrator samples.

For instructions about how to perform the calibration procedure, refer to the online help.

Performing Quality Control

For quality control of the Atellica IM Fol assay, use an appropriate quality control material of known analyte concentration with at least 2 levels (low and high) at least once during each day that samples are analyzed. For assistance in identifying a quality control material, refer to *Atellica® IM Quality Control Material Supplement* available on siemens-healthineers.com.

Additional quality control material can be used at the discretion of the laboratory. Use the quality control material in accordance with the quality control instructions for use.

In addition, perform quality control:

- Following a valid calibration
- With use of a new lot of reagent
- When troubleshooting test results that do not match clinical conditions or symptoms

Follow government regulations or accreditation requirements for quality control frequency. Individual laboratory quality control programs and procedures may require more frequent quality control testing.

Acceptable performance is achieved when the analyte values obtained are within the expected control interval for the system, as indicated by the manufacturer of the control material or within the interval determined by an internal laboratory quality control procedure.

Follow your laboratory's quality control procedures if the results obtained do not fall within the acceptable limits. For information about entering quality control definitions, refer to the system online help.

Taking Corrective Action

If the quality control results do not fall within the assigned values, do not report results. Perform corrective actions in accordance with established laboratory protocol. For suggested protocol, refer to the online help.

Results

Calculation of Results

The system determines the result using the calculation scheme described in the online help. The system reports results in ng/mL (common units) or nmol/L (SI units), depending on the units defined when setting up the assay.

Conversion formula: 1 ng/mL = 2.265 nmol/L

For information about results outside the specified measuring interval, refer to *Measuring Interval*.

Atellica IM Test Options

The Atellica IM FoI assay can be used for the following test options:

- FoI = system test generated whole blood folate concentration
- rFOL = predefined ratio test on the system for system-generated hemolysate folate value
- rbcFOL = predefined ratio test on the system for system-generated hemolysate RBC folate value (requires off-system hematocrit value)

  Note For information about creating the off-system hematocrit test, refer to the online help.
- Manually calculated RBC Folate value using the rFOL predefined ratio value generated by the system for the hemolysate and the off-system hematocrit value
- Corrected Red Blood Cell Folate = system or manually calculated corrected RBC Folate

For information about entering the off-system and ratio test definitions, refer to the online help.

Manual Calculation for Red Blood Cell Folate

Use this procedure to manually calculate RBC folate values using an off-system hematocrit value and the result from the rFOL test.

1. Prepare the RBC hemolysate as described in *Preparing the Red Blood Cell Hemolysate*.
2. Order the rFOL predefined ratio test.

   Note The rFOL predefined ratio test must use RBC hemolysate sample. Do not use other sample types.
3. Multiply the folate result for the hemolysate by 21 (a 1:21 dilution was made when preparing the RBC hemolysate). This value represents the folate concentration of whole blood in ng/mL.
4. Divide this result by the hematocrit, and multiply by 100 to adjust for the hematocrit, which is a percentage.

$$\text{RBC folate (ng/mL)} = \frac{\text{Folate result for hemolysate, (ng/mL) x 21}}{\text{hematocrit}} \times 100$$

Example:
Hemolysate folate value = 5.7 ng/mL
Hematocrit = 43

$$\text{RBC folate (ng/mL)} = \frac{5.7 \times 21}{43} \times 100 = 278$$

Red Blood Cell Folate using the predefined system ratio test (rbcFOL)

1. Prepare the RBC hemolysate as described in *Preparing the Red Blood Cell Hemolysate*.
2. Order rbcFOL from the list of predefined ratio tests.

   Note The rbcFOL predefined ratio test must use RBC hemolysate sample. Do not use other sample types.
3. Manually enter the hematocrit value for the sample.
4. The system will display the calculated red blood cell folate value.

Corrected Red Blood Cell Folate

In most cases the serum folate values are very small compared to red blood cell folate values. However, occasionally serum folate values can be elevated. If the serum folate value is high and the red blood cell folate concentration is low, calculate the corrected RBC folate value according to the following equation:

$$\text{Corrected RBC folate (ng/mL)} = \underset{\text{(ng/mL)}}{\text{RBC folate}} - \underset{\text{(ng/mL)}}{\text{serum folate}} \left[\frac{(100 - \text{hematocrit})}{\text{hematocrit}}\right]$$

Example:
RBC folate = 210 ng/mL
Serum folate = 22 ng/mL
Hematocrit of the patient = 41

$$\text{Corrected RBC folate (ng/mL)} = 210 - 22 \left[\frac{(100 - 41)}{41}\right] = 210 - 32 = 178$$

For information about entering the off-system and ratio test definitions, refer to the online help.

Dilutions

The assay measuring interval for serum is 0.35–24.00 ng/mL (0.79–54.36 nmol/L). For information about dilution options used to extend the reportable measuring interval up to 48.00 ng/mL (108.72 nmol/L), refer to the online help.

Serum samples with folate levels > 24.00 ng/mL (> 54.36 nmol/L) must be diluted and retested to obtain accurate results.

Note Do not dilute the RBC hemolysate.

For automated dilutions, ensure that Atellica IM Fol DIL is loaded on the system. Ensure that sufficient sample volume is available to perform the dilution and that the appropriate dilution factor is selected when scheduling the test, as indicated in the table below.

For automatic dilutions, enter a dilution setpoint ≤ 24.00 ng/mL (≤ 54.36 nmol/L).

Sample	Dilution	Sample Volume (µL)
Serum	1:2	100

Interpretation of Results

Results of this assay should always be interpreted in conjunction with the patient's medical history, clinical presentation, and other findings.

Limitations

The following information pertains to limitations of the assay:

- Hemolysis significantly increases folate values in serum due to the high folate concentrations found in red blood cells.
- Methotrexate and leucovorin interfere with folate measurement because these drugs cross-react with folate binding proteins.

Expected Values

The reagent formulations used on the Atellica IM Analyzer are the same as those used on the ADVIA Centaur® system. Expected values were established using the ADVIA Centaur system and confirmed by assay comparison. Refer to *Assay Comparison*.

To determine the reference intervals for serum and RBC folate, data were obtained using 370 and 286 samples, respectively. For sample results in the indeterminate range [3.38–5.38 ng/mL (7.64–12.19 nmol/L)], clinical results and other diagnostic protocols should supplement folate results.

Category	N[a]	Median (ng/mL)	Range (ng/mL)	Median (nmol/L)	Range (nmol/L)
Serum folate					
Deficient[c]	65	1.54	0.35–3.37	3.49	0.79–7.63
Indeterminate[d]			3.38–5.38		7.64–12.19
Normal	305	12.51	> 5.38[b]	28.34	> 12.19
RBC folate					
Normal	286	425	280–791	963	634–1792

[a] Number of samples.
[b] Inner 97.5% of the distribution of apparently healthy individuals.
[c] Diagnosed by bone and/or peripheral blood smear pathology and other criteria including:
- megaloblastic anemia
- folate-deficient diet
- malabsorption
- alcoholism
- Tropical Sprue
- abnormal blood parameters including mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and hematocrit (HCT)

[d] Range between deficient and normal range.

Laboratories should consider these expected values as guidelines only. The data were obtained on apparently healthy males and females from the United States. Because of population demographic factors, diet, and assay methods, each laboratory should determine its own expected values for the diagnostic evaluation of patient results.[13]

Performance Characteristics

The reagent formulations used on the Atellica IM Analyzer are the same as those used on the ADVIA Centaur system. Some performance characteristics for the Atellica IM assay were established using the ADVIA Centaur system.

Measuring Interval

The Atellica IM Fol assay provides results from 0.35–24.00 ng/mL (0.79–54.36 nmol/L). The lower end of the measuring interval is defined by the analytical sensitivity. Report results below the measuring interval as < 0.35 ng/mL (< 0.79 nmol/L). When sample results exceed the measuring interval, refer to *Dilutions*.

Detection Capability

Detection capability was determined in accordance with CLSI Document EP17-A2.[14] The assay is designed to have an analytical sensitivity of ≤ 0.35 ng/mL (0.79 nmol/L), a limit of blank (LoB) ≤ 0.40 ng/mL (0.91 nmol/L), and a limit of detection (LoD) ≤ 0.70 ng/mL (1.59 nmol/L).

Representative detection capability data are shown below. Assay results obtained at individual laboratories may vary from the data presented.

Analytical sensitivity is defined as the concentration of folate that corresponds to the RLUs that are 2 standard deviations less than the mean RLUs of 20 replicate determinations of the folate zero standard. This response is an estimate of the minimum detectable concentration with 95% confidence. The analytical sensitivity for the Atellica IM Fol assay is < 0.01 ng/mL (< 0.02 nmol/L).

The LoB corresponds to the highest measurement result that is likely to be observed for a blank sample. The LoB of the Atellica IM Fol assay is 0.19 ng/mL (0.43 nmol/L).

The LoD corresponds to the lowest concentration of folate that can be detected with a probability of 95%. The LoD for the Atellica IM Fol assay is 0.38 ng/mL (0.86 nmol/L), and was determined using 540 determinations, with 480 blank and 60 low-level replicates, and an LoB of 0.19 ng/mL (0.43 nmol/L).

Precision

Precision was determined in accordance with CLSI Document EP05-A3.[15] Samples were assayed on an Atellica IM Analyzer in duplicate in 2 runs per day for 20 days. The assay was designed to have within-laboratory precision for serum ≤ 0.21 ng/mL (0.48 nmol/L) SD for samples < 1.80 ng/mL (4.08 nmol/L) and ≤ 11% CV for samples from 1.90–24.00 ng/mL (4.30–54.36 nmol/L).

The assay was designed to have within-laboratory precision for red blood cell hemolysate ≤ 14.50 ng/mL (32.84 nmol/L) SD for samples < 130.00 ng/mL (294.45 nmol/L) and ≤ 11% CV for samples from 131.00–580.00 ng/mL (296.72–1313.70 nmol/L).

	Mean		Repeatability			Within-Laboratory Precision		
			SD[a]		CV[b]	SD		CV
Sample Type	(ng/mL)	(nmol/L)	(ng/mL)	(nmol/L)	(%)	(ng/mL)	(nmol/L)	(%)
Serum A	1.42	3.22	0.05	0.11	N/A[c]	0.08	0.18	N/A
Serum B	4.13	9.35	0.10	0.23	2.4	0.24	0.54	5.9
Serum C	6.19	14.02	0.18	0.41	2.9	0.36	0.82	5.9
Serum D	9.23	20.91	0.24	0.54	2.6	0.6	1.36	6.5
Serum E	21.91	49.63	0.72	1.63	3.3	1.57	3.56	7.1

	Mean		Repeatability			Within-Laboratory Precision		
			SD[a]		CV[b]	SD		CV
Sample Type	**(ng/mL)**	**(nmol/L)**	**(ng/mL)**	**(nmol/L)**	**(%)**	**(ng/mL)**	**(nmol/L)**	**(%)**
Serum Control 1	2.82	6.39	0.09	0.20	3.3	0.17	0.39	6.1
Serum Control 2	5.43	12.30	0.14	0.32	2.5	0.35	0.79	6.4
Red Blood Cell Sample A	75.23	170.40	4.58	10.37	N/A	6.57	14.88	N/A
Red Blood Cell Sample B	133.64	302.69	4.42	10.01	3.3	11.25	25.48	8.4
Red Blood Cell Sample C	343.79	778.68	9.86	22.33	2.9	20.05	45.41	5.8
Red Blood Cell Sample D	545.81	1236.26	19.40	43.94	3.6	35.29	79.93	6.5
Red Blood Cell Sample E	882.20	1998.18	45.80	103.74	5.2	63.13	142.99	7.2
Red Blood Cell Sample F	1258.33	2850.12	88.55	200.57	7.0	97.66	221.20	7.8
Whole Blood Control 1	57.52	130.28	2.69	6.09	N/A	5.5	12.46	N/A
Whole Blood Control 2	238.39	539.95	6.53	14.79	2.7	16.9	38.28	7.1

[a] Standard deviation.
[b] Coefficient of variation.
[c] Not applicable.

Based on internal testing on the Atellica IM Analyzer, the overall reproducibility is estimated to be ≤ 20% CV for samples tested and includes multiple reagent lots, instruments, days, and replicates. Performance of the assay at individual laboratories may vary.

Collection Tube Comparison

Red Cell Folate

The red cell folate values from 13 donors were determined for blood collected in both EDTA and heparin tubes. Samples drawn in heparin tubes were on average 10% higher than samples drawn in EDTA tubes.

	EDTA		Heparin	
	(ng/mL)	**(nmol/L)**	**(ng/mL)**	**(nmol/L)**
Mean	594	1345	652	1477
Range	421–1098	954–2487	500–1179	1162–2670

Assay Comparison

The Atellica IM Fol assay is designed to have a correlation coefficient of ≥ 0.95 and a slope of 1.0 ±0.1 for serum when compared to the ADVIA Centaur Folate assay. Assay comparison was determined using the weighted Deming regression model in accordance with CLSI Document EP09-A3.[16] The following results were obtained:

Specimen	Comparative Assay (x)	Regression Equation	Sample Interval	N[a]	r[b]
Serum	ADVIA Centaur Folate	y = 0.94x - 0.01 ng/mL (y = 0.94x - 0.02 nmol/L)	0.64–22.78 ng/mL (1.45–51.60 nmol/L)	105	0.99
RBC hemolysate	ADVIA Centaur Folate	y = 0.93x + 25.89 ng/mL (y = 0.93x + 58.64 nmol/L)	181.65–1343.39 ng/mL (411.44–3042.78 nmol/L)	120	0.94

[a] Number of samples tested.

[b] Correlation coefficient.

Agreement of the assays may vary depending on the study design, comparative assay, and sample population used. Assay results obtained at individual laboratories may vary from the data presented.

Interferences

The Atellica IM Fol assay is designed to have ≤ 10% interference for bilirubin and lipemia. Interfering substances at the levels indicated in the table below were tested in accordance with CLSI Document EP07-A2[17] using the Atellica IM Analyzer.

Bias is the difference in the results between the control sample (does not contain the interferent) and the test sample (contains the interferent) expressed in percent. Analyte results should not be corrected based on this bias.

Substance	Substance Test Concentration Common Units (SI Units)	Analyte Concentration ng/mL (nmol/L)	Bias (%)
Bilirubin, conjugated	20 mg/dL (341 µmol/L)	3.8 (8.61)	0.7
	20 mg/dL (341 µmol/L)	9.5 (21.52)	9.1
Bilirubin, unconjugated	20 mg/dL (341 µmol/L)	3.6 (8.15)	-5.3
	20 mg/dL (341 µmol/L)	9.8 (22.20)	-1.5
Lipemia (Intralipid®)	2000 mg/dL (22.7 mmol/L)	5.0 (11.33)	2.0
	2000 mg/dL (22.7 mmol/L)	8.8 (19.93)	3.0

Biotin was added to serum samples containing different concentrations of folate. These samples were tested against an appropriate control and the observed bias is presented in the following table:

Analyte Concentration ng/mL (nmol/L)	Biotin Test Level in Serum (ng/mL)								
	0	1	5	19	38	75	150	300	600
	% Bias								
9.23 (20.91)	—	1.4	-1.1	6.9	4.7	20.2	> MI[a]	> MI	> MI
17.37 (39.34)	—	1.3	1.3	3.6	8.3	35.3	> MI	> MI	> MI

[a] Measuring Interval

In addition, biotin was added to whole blood samples containing different concentrations of folate. These samples were tested against an appropriate control and the observed bias is presented in the following table:

Analyte Concentration ng/mL (nmol/L)	Biotin Test Level in Whole Blood (ng/mL)								
	0	1	5	19	38	75	150	300	600
	% Bias								
13.19 (29.88)	—	-6.8	-8.1	-8.5	-5.6	-3.4	-1.1	47.9	65.8
18.90 (42.81)	—	0.1	4.4	-0.2	4.3	7.6	12.4	> MI[a]	> MI

[a] Measuring Interval

Specimens that contain biotin at a concentration of 50 ng/mL (in serum) or 75 ng/mL (in whole blood) demonstrate a less than or equal to 10% change in results. Biotin concentrations greater than these may lead to falsely elevated results for patient samples.

The recommended adult daily dietary intake for biotin is 30 µg/day. Over the counter dietary supplements promoted for use in hair, skin, and nail health may contain 5–100 mg of biotin, with recommendations to take multiple pills per day. Pharmacokinetic studies in healthy adults have shown that, in subjects ingesting 5 mg, 10 mg, and 20 mg of biotin, serum concentrations of biotin can reach up to 73 ng/mL, 141 ng/mL, and 355 ng/mL, respectively.[18] Subjects who take up to 300 mg of biotin per day may have plasma biotin levels as high as 1160 ng/mL.[19]

Assay results obtained at individual laboratories may vary from the data presented.

Dilution Recovery

Five human serum samples in the range of 12.42–22.36 ng/mL (28.13–50.65 nmol/L) of folate were diluted 1:2 with Atellica IM Fol DIL and assayed for recovery and parallelism. The recoveries ranged from 90.0%–99.4% with a mean of 95.3%.

Sample	Dilution	Observed (ng/mL)	Expected (ng/mL)	Observed (nmol/L)	Expected (nmol/L)	Recovery (%)
1	—	22.36	—	50.65	—	—
	1:2	10.06	11.18	22.79	25.32	90.0
2	—	16.82	—	38.10	—	—
	1:2	8.21	8.41	18.60	19.05	97.6
3	—	12.42	—	28.13	—	—
	1:2	6.03	6.21	13.66	14.07	97.1
4	—	15.50	—	35.11	—	—
	1:2	7.17	7.75	16.24	17.55	92.5
5	—	20.90	—	47.34	—	—
	1:2	10.39	10.45	23.53	23.67	99.4
Mean						**95.3**

Results were established using the Atellica IM Analyzer. Assay results obtained at individual laboratories may vary from the data presented.

Spiking Recovery

Varying amounts of folate were added to 5 samples with endogenous folate levels of 1.05–1.35 ng/mL (2.38–3.06 nmol/L). The recoveries ranged from 87%–116% with a mean of 104%.

Sample	Amount Added (ng/mL)	Observed (ng/mL)	Amount Added (nmol/L)	Observed (nmol/L)	Recovery (%)
1	0.00	1.05	0.00	2.38	—
	3.57	4.65	8.09	10.53	108
	7.14	8.50	16.17	19.25	112
	10.70	12.20	24.24	27.63	115
	16.70	16.40	37.83	37.15	109
	19.00	18.30	43.04	41.45	108
	Mean				110
2	0.00	1.11	0.00	2.51	—
	3.57	4.65	8.09	10.53	105
	7.14	8.40	16.17	19.03	111
	10.70	12.30	24.24	27.86	116
	16.70	15.50	37.83	35.11	101
	19.00	19.60	43.04	44.39	116
	Mean				109
3	0.00	1.30	0.00	2.94	—
	3.57	4.45	8.09	10.08	95
	7.14	7.80	16.17	17.67	101
	10.70	11.40	24.24	25.82	106
	16.70	13.60	37.83	30.80	87
	19.00	17.10	43.04	38.73	99
	Mean				97
4	0.00	1.35	0.00	3.06	—
	2.98	3.97	6.75	8.99	94
	5.95	6.74	13.48	15.27	99
	8.90	9.40	20.16	21.29	99
	11.90	11.60	26.95	26.27	99
	14.90	14.50	33.75	32.84	103
	Mean				99
5	0.00	1.35	0.00	3.06	—
	2.98	4.11	6.75	9.31	101

Sample	Amount Added (ng/mL)	Observed (ng/mL)	Amount Added (nmol/L)	Observed (nmol/L)	Recovery (%)
	5.95	7.21	13.48	16.33	106
	8.90	10.10	20.16	22.88	108
	11.90	11.90	26.95	26.95	100
	14.90	14.90	33.75	33.75	105
	Mean				104
Mean					**104**

Results were established using the Atellica IM Analyzer. Assay results obtained at individual laboratories may vary from the data presented.

Standardization

The Atellica IM Fol assay is traceable to an internal standard manufactured using highly purified material (N-5-methyl tetrahydrofolate). Assigned values for calibrators are traceable to this standardization.

Technical Assistance

According to EU regulation 2017/746, any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority of the EU Member State in which the user and/or patient is established.

For customer support, contact your local technical support provider or distributor.

siemens-healthineers.com

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Definition of Symbols

The following symbols may appear on the product labeling:

Symbol	Symbol Title	Source	Symbol	Symbol Title	Source
	Manufacturer	5.1.1[a]		Authorized representative in the European Community	5.1.2[a]
	Use-by date	5.1.4[a]		Authorized representative in Switzerland	Proprietary
	Catalog number	5.1.6[a]		Batch code	5.1.5[a]
	Consult Instructions for Use	5.4.3[a]		Contains sufficient for <n> tests	5.5.5[a]
	Internet URL address to access the electronic instructions for use	Proprietary		Version of Instructions for Use	Proprietary
	*In vitro* diagnostic medical device	5.5.1[a]		Revision	Proprietary

Symbol	Symbol Title	Source	Symbol	Symbol Title	Source
RxOnly	Prescription device (US only)	FDA[b]	UDI	Unique Device Identifier	5.7.10[c]
CE XXXX	CE Marking with Notified Body	EU IVDR[d]	CE	CE Marking	EU IVDR[d]
	Temperature limit	5.3.7[a]		Keep away from sunlight	5.3.2[a]
	Upper limit of temperature	5.3.6[a]		Lower limit of temperature	5.3.5[a]
	Do not re-use	5.4.2[a]		Do not freeze	Proprietary
	Recycle	1135[e]		This way up	0623[e]
	Biological risks	5.4.1[a]		Caution	5.4.4[a]
UNITS C	Common Units	Proprietary	UNITS SI	International System of Units	Proprietary
YYYY-MM-DD	Date format (year-month-day)	N/A	YYYY-MM	Date format (year-month)	N/A
	Document face up[f]	1952[e]		Handheld barcode scanner	Proprietary
	Target	Proprietary		Mixing of substances	5657[g]
CHECKSUM	Variable hexadecimal number that ensures the Master Curve and Calibrator definition values entered are valid.	Proprietary		Interval	Proprietary
MATERIAL ID	Unique material identification number	Proprietary	MATERIAL	Material	Proprietary
CONTROL TYPE	Type of control	Proprietary	CONTROL NAME	Name of control	Proprietary
CONTROL LOT VAL	Quality control lot value	Proprietary	CAL LOT VAL	Calibrator lot value	Proprietary

[a] International Standard Organization (ISO). ISO 15223-1 Medical Devices- Symbols to be used with medical device labels, labelling and information to be supplied.
[b] Federal Register. Vol. 81, No 115. Wednesday, June 15, 2016. Rules and Regulations: 38911.
[c] ISO 15223-1:2020-04
[d] IVDR REGULATION (EU) 2017/746
[e] International Standard Organization (ISO). ISO 7000 Graphical symbols for use on equipment.
[f] Indicates Assay-eNote
[g] International Electrotechnical Commission (IEC). IEC 60417-1 Graphical symbols for use on equipment – Part 1: Overview and Application

Legal Information

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SIEMENS Healthineers

Folate (Fol)

Current Revision and Date[a]	Rev. 05, 2024-05	
Product Name	Atellica IM Folate (Fol)	REF 10995572 (140 tests) REF 10995573 (700 tests)
Abbreviated Product Name	Atellica IM Fol	
Test Name/ID	Fol FolSerum	
Systems	Atellica CI Analyzer	
Materials Required but Not Provided	Atellica IM Fol DTT/REL	REF 10995576
	Atellica IM APW1	REF 10995458
Optional Materials	Atellica IM RBC Fol	REF 10995440
	Atellica IM Fol DIL	REF 10995574
	Atellica IM Fol MCM	REF 10995575
Specimen Types	Serum, heparinized whole blood	
Sample Volume	100 µL	
Measuring Interval	0.35–24.00 ng/mL (0.79–54.36 nmol/L)	

[a] A vertical bar in the page margin indicates technical content that differs from the previous version.

CE 0197

Intended Use

The Atellica® IM Folate (Fol) assay is for *in vitro* diagnostic use in the quantitative determination of folate in human serum and red blood cells using the Atellica® CI Analyzer.

Summary and Explanation

Folates are compounds of pteroylglutamic acid (PGA) that function as coenzymes in metabolic reactions involving the transfer of single-carbon units from a donor to a recipient compound. Folate, with vitamin B_{12}, is essential for DNA synthesis, which is required for normal red blood cell maturation.[1] Humans obtain folate from dietary sources including fruits, green and leafy vegetables, yeast, and organ meats.[2] Folate is absorbed through the small intestine and stored in the liver. Measurements of folate are used as an aid in the diagnosis and treatment of folate deficiency.[3]

Low folate intake, malabsorption as a result of gastrointestinal diseases, pregnancy, and drugs such as phenytoin are causes of folate deficiency.[4] Folate deficiency is also associated with chronic alcoholism.[5] Folate and vitamin B_{12} deficiency impair DNA synthesis, causing macrocytic anemias. These anemias are characterized by abnormal maturation of red blood cell precursors in the bone marrow, the presence of megaloblasts, and decreased red blood cell survival.[1]

Since both folate and vitamin B_{12} deficiency can cause macrocytic anemia, appropriate treatment depends on the differential diagnosis of the deficiency. Serum folate measurement provides an early index of folate status.[2] However, folate is much more concentrated in red blood cells than in serum so the red blood cell folate measurement more closely reflects tissue stores.[5] Red blood cell folate concentration is considered the most reliable indicator of folate status.[2]

Principles of the Procedure

The Atellica IM Fol assay is a competitive immunoassay using direct chemiluminescent technology. Folate in the patient sample competes with acridinium-ester-labeled folate in the Lite Reagent for a limited amount of biotin-labeled folate binding protein. Biotin-labeled folate binding protein binds to avidin that is covalently coupled to paramagnetic particles in the Solid Phase. In the Atellica IM Fol assay, the sample is pretreated to release the folate from endogenous binding proteins in the sample.

An inverse relationship exists between the amount of folate present in the patient sample and the amount of relative light units (RLUs) detected by the system.

Reagents

Material Description	Storage	Stability[a]
Atellica IM Fol ReadyPack® primary reagent pack **Lite Reagent** 9.1 mL/reagent pack Folate labeled with acridinium ester (~9.8 ng/mL) in buffer; bovine serum albumin; sodium azide (0.1%); preservatives **Solid Phase** 18.2 mL/reagent pack Purified avidin (~20 µg/mL) covalently coupled to paramagnetic particles in buffer; human serum albumin; preservatives **Folate Binding Protein** 9.1 mL/reagent pack Purified folate binding protein (~1.0 µg/mL) covalently coupled to biotin in buffer; bovine serum albumin; preservatives	Unopened at 2–8°C Onboard	Until expiration date on product 7 days

Material Description	Storage	Stability[a]
Atellica IM Fol CAL 3.0 mL/vial; lyophilized After reconstitution, low or high levels of N-5-methyltetrahydrofolic acid; buffer; human serum albumin; sodium azide (< 0.1%); preservatives	Lyophilized at 2–8°C	Until expiration date on product
	Reconstituted at 2–8°C	7 days
	Reconstituted at room temperature	8 hours
	Reconstituted at ≤ -20°C	28 days
Atellica IM Fol DTT/Releasing Agent[b] **DTT** 8.0 mL/vial Dithiothreitol (~95 mg/mL in liquid form) **Releasing Agent** 4.0 mL/vial Sodium hydroxide (~1.1 N)	At 2–8°C	Until expiration date on product
	Onboard in an ancillary reagent pack[c]	108 hours
Atellica IM RBC Fol[d, e] **RBC Folate Ascorbic Acid** Lyophilized ascorbic acid (~0.30 g/vial) **RBC Folate Ascorbic Acid Diluent** 30.0 mL/vial Bovine serum albumin; buffer; sodium azide (< 0.1%); preservatives	Unopened at 2–8°C	Until expiration date on product
	Reconstituted at 2–8°C	30 days
Atellica IM Fol DIL ReadyPack ancillary reagent pack[e] 10.0 mL/reagent pack Human plasma; sodium azide (< 0.1%); preservatives	Unopened at 2–8°C	Until expiration date on product
	Onboard	28 days
Atellica IM APW1 ReadyPack ancillary reagent pack[c] 25.0 mL/pack 0.4 N sodium hydroxide	Unopened at 2–8°C	Until expiration date on product
	Onboard	14 days

[a] Refer to *Storage and Stability*.
[b] Refer to *Materials Required but Not Provided*.
[c] Refer to *Preparing the Reagents*.
[d] Refer to *Optional Materials*.
[e] Refer to *Preparing the Red Blood Cell Hemolysate* in the *Preparing the Samples* section.

Warnings and Precautions

For *in vitro* diagnostic use.

For Professional Use.

CAUTION

Federal (USA) law restricts this device to sale by or on the order of a licensed healthcare professional.

Safety data sheets (SDS) available on siemens-healthineers.com.

H290, H314
P234, P264, P280, P301+P330+P331, P303+P361+P353, P310, P305+P351+P338, P390, P501

Danger!
May be corrosive to metals. Causes severe skin burns and eye damage. Keep only in original container. Wash hands thoroughly after handling. Wear protective gloves/protective clothing/eye protection/face protection. IF SWALLOWED: rinse mouth. Do NOT induce vomiting. IF ON SKIN (or hair): Remove/Take off immediately all contaminated clothing. Rinse skin with water/shower. Immediately call a POISON CENTER or doctor/physician. IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Absorb spillage to prevent material damage. Dispose of contents and container in accordance with all local, regional, and national regulations.
Contains: sodium hydroxide (Atellica IM Fol Releasing Agent)

H290, H319, H315
P234, P264, P280, P337+P313, P390, P501

Warning!
May be corrosive to metals. Causes serious eye irritation. Causes skin irritation.
Keep only in original container. Wash hands thoroughly after handling. Wear protective gloves/protective clothing/eye protection/face protection. If eye irritation persists: Get medical advice/attention. Absorb spillage to prevent material damage. Dispose of contents and container in accordance with all local, regional, and national regulations.
Contains: sodium hydroxide (Atellica IM APW1)

H412
P273, P501

Harmful to aquatic life with long lasting effects.
Avoid release to the environment. Dispose of contents and container in accordance with all local, regional, and national regulations.
Contains: sodium azide (in Atellica IM Fol CAL)

H360F, H360D
P201, P280, P308+P313

Danger!
May damage fertility. May damage the unborn child.
Obtain special instructions before use. Wear protective gloves/protective clothing/eye protection/face protection. IF exposed or concerned: Get medical advice/attention.
Contains: disodium tetraborate decahydrate (Atellica IM FOL Solid phase and FOL Binding Protein)

Warning! Potential Biohazard

Contains human source material.

No known test method can ensure that products derived from human source materials will not transmit infection. These materials should be handled using good laboratory practices and universal precautions.[6-8]

CAUTION

This device contains material of animal origin and should be handled as a potential carrier and transmitter of disease.

Contains sodium azide as a preservative. Sodium azide can react with copper or lead plumbing to form explosive metal azides. On disposal, flush reagents with a large volume of water to prevent buildup of azides. Disposal into drain systems must be in compliance with prevailing regulatory requirements.

Dispose of hazardous or biologically contaminated materials according to the practices of your institution. Discard all materials in a safe and acceptable manner and in compliance with prevailing regulatory requirements.

Note For information about reagent preparation, refer to *Preparing the Reagents* in the *Procedure* section.

Note For information about calibrator preparation, refer to *Preparing the Calibrators*.

Storage and Stability

Store reagents in an upright position. Protect the product from heat and light sources. Unopened reagents are stable until the expiration date on the product when stored at 2–8°C.

Store calibrators in an upright position. Lyophilized calibrators are stable until the expiration date on the product when stored at 2–8°C. Reconstituted calibrators are stable for 7 days at 2–8°C or 8 hours at room temperature. Freeze reconstituted product at ≤ -20°C for up to 28 days.

Store Atellica IM Fol DTT/Releasing Agent in an upright position. Unopened Atellica IM Fol DTT/Releasing Agent is stable until the expiration date on the product when stored at 2–8°C.

Store Atellica IM RBC Fol in an upright position. Unopened Atellica IM RBC Fol is stable until the expiration date on the product when stored at 2–8°C. Reconstituted material is stable for 30 days at 2–8°C.

Store Atellica IM Fol DIL in an upright position. Unopened Atellica IM Fol DIL is stable until the expiration date on the product when stored at 2–8°C.

Store Atellica IM APW1 in an upright position. Unopened Atellica IM APW1 is stable until the expiration date on the product when stored at 2–8°C.

Do not use products beyond the expiration date printed on the product labeling.

Onboard Stability

Discard products at the end of the onboard stability interval. Do not use products beyond the expiration date printed on the product labeling.

For information about product onboard stability, refer to *Reagents*.

Specimen Collection and Handling

Serum and red blood cells are the recommended sample types for this assay.

Note Folates are light-sensitive. Minimize exposure to light during sample handling and storage.[9]

Collecting the Specimen

- Observe universal precautions when collecting specimens. Handle all specimens as if they are capable of transmitting disease.[8]
- Follow recommended procedures for collection of diagnostic blood specimens by venipuncture.[10]
- Follow the instructions provided with your specimen collection device for use and processing.[11]
- Allow blood specimens to clot completely before centrifugation.[12]
- Keep tubes capped at all times.[12]

Storing the Specimen

- Store serum samples at room temperature for no longer than 8 hours.
- Tightly cap and refrigerate specimens at 2–8°C if the assay is not completed within 8 hours.
- Freeze serum samples at ≤ -20°C if the assay is not completed within 48 hours.
- Freeze serum samples only 1 time and mix thoroughly after thawing. Frozen specimens can remain frozen up to 30 days. Do not store in a frost-free freezer. If serum samples will be stored for longer than 30 days, then they must be frozen at ≤ -80°C.
- If testing is not done within 24 hours for whole blood specimens, determine the hematocrit and freeze the whole blood specimen or hemolysate. Frozen whole blood specimens can be stored at -20°C for up to 2 months. Sample hemolysates prepared with the reconstituted RBC Folate Ascorbic Acid can be stored at -20°C for up to 3 months. Do not store whole blood specimens or hemolysates in a frost-free freezer. Freeze specimens only 1 time and mix thoroughly after thawing.

The handling and storage information provided here is based on data or references maintained by the manufacturer. It is the responsibility of the individual laboratory to use all available references and/or its own studies when establishing alternate stability criteria to meet specific needs.

Transporting the Specimen

Package and label specimens for shipment in compliance with applicable federal and international regulations covering the transport of clinical specimens and etiological agents.

Preparing the Samples

This assay requires 100 µL of sample for a single determination. This volume does not include the unusable volume in the sample container or the additional volume required when performing duplicates or other tests on the same sample. For information about determining the minimum required volume, refer to the system online help.

The sample volume required to perform onboard dilution differs from the sample volume required to perform a single determination. Refer to *Dilutions*.

Note Do not use specimens with apparent contamination.

Before placing samples on the system, ensure that samples are free of:

- Bubbles or foam.
- Fibrin or other particulate matter.

Note Remove particulates by centrifugation according to CLSI guidance and the collection device manufacturer's recommendations.[12]

Note For a complete list of appropriate sample containers, refer to the system online help.

Preparing the Red Blood Cell Hemolysate

Note Folates are light sensitive. Minimize exposure to light during sample handling and storage.[9]

1. Reconstitute the RBC Folate Ascorbic Acid by adding the contents of 1 vial of RBC Folate Ascorbic Acid Diluent to the lyophilized RBC Folate Ascorbic Acid.
2. Let the reconstituted mixture stand at room temperature for 15 minutes and mix by inverting the bottle occasionally.
3. Collect the sample in a tube containing heparin or EDTA.
4. Invert the sample several times to mix.

5. Determine and record the hematocrit.
6. Dispense 1.0 mL of reconstituted RBC Folate Ascorbic Acid into a test tube or sample cup.
7. Add 50 µL of the sample into the RBC Folate Ascorbic Acid.
8. Cap and invert the tube several times or vortex gently to mix. Avoid foaming.
9. Let the hemolysate stand protected from light, at room temperature, **for a minimum of 90 minutes, but less than 3 hours**.
10. **Do not mix the hemolysate again before placing the sample on the system.**

Note Do not dilute the RBC hemolysate. Freeze the hemolysate at ≤ -20°C immediately if testing cannot be completed within 4 hours from the time you finish preparing the hemolysate. Hemolysates can be stored at ≤ -20°C for up to 3 months. Do not store in a frost-free freezer.

If the hemolysate is frozen, thaw the hemolysate and mix it by inverting the tube several times. Let the hemolysate stand for 30 minutes at room temperature before testing. Test the hemolysate within 3 hours after thawing.

Procedure

Materials Provided

The following materials are provided:

REF	Contents	Number of Tests
10995572	1 ReadyPack primary reagent pack containing Atellica IM Fol Lite Reagent, Solid Phase, and Folate Binding Protein Fol (whole blood) master curve and test definition MC TDEF FolSR (serum) master curve and test definition MC TDEF 2 vials Atellica IM Fol CAL low calibrator CAL L 2 vials Atellica IM Fol CAL high calibrator CAL H Atellica IM Fol CAL calibrator lot-specific value sheet CAL LOT VAL	140
10995573	5 ReadyPack primary reagent packs containing Atellica IM Fol Lite Reagent, Solid Phase, and Folate Binding Protein Fol (whole blood) master curve and test definition MC TDEF FolSR (serum) master curve and test definition MC TDEF 2 vials Atellica IM Fol CAL low calibrator CAL L 2 vials Atellica IM Fol CAL high calibrator CAL H Atellica IM Fol CAL calibrator lot-specific value sheet CAL LOT VAL	700

Materials Required but Not Provided

The following materials are required to perform this assay, but are not provided:

REF	Description	
	Atellica CI Analyzer[a]	
10995576	Atellica IM Fol DTT/REL (releasing agent)	3 x 8.0 mL/vial DTT 3 x 4.0 mL/vial Releasing Agent REL 3 empty ReadyPack ancillary reagent packs
10995458	Atellica IM APW1 (probe wash)	2 ReadyPack ancillary reagent packs containing 25.0 mL/pack WASH

[a] Additional system fluids are required to operate the system. For system fluid instructions for use, refer to the Document Library.

Optional Materials

The following materials may be used to perform this assay, but are not provided:

REF	Description	
10995440	Atellica IM RBC Fol (ascorbic acid/diluent)	4 x 0.30 g/vial Ascorbic Acid ASCORBIC ACID 4 x 30.0 mL/vial Ascorbic Acid Diluent ASCORBIC ACID DIL
10995574	Atellica IM Fol DIL (diluent)	2 ReadyPack ancillary reagent packs containing 10.0 mL/pack DIL
10995575	Atellica IM Fol MCM (master curve material)	6 x 1.0 mL levels of master curve material MCM

Assay Procedure

The system automatically performs the following steps:

1. Dispenses 100 µL of sample into a cuvette.
2. Dispenses 35 µL of DTT/Releasing Agent, then incubates for 5 minutes at 37°C.
3. Dispenses 65 µL of Folate Binding Protein and 130 µL of Solid Phase, then incubates for 5 minutes at 37°C.
4. Dispenses 65 µL of Lite Reagent, then incubates for 3 minutes at 37°C.
5. Separates, aspirates, then washes the cuvettes with Atellica IM Wash.
6. Dispenses 300 µL each of Atellica IM Acid and Atellica IM Base to initiate the chemiluminescent reaction.
7. Reports results.

Preparing the Reagents

All reagents are liquid and ready to use, with the exception of the Atellica IM Fol DTT/Releasing Agent. Before loading primary reagent packs onto the system, mix them by hand and visually inspect the bottom of the reagent pack to ensure that all particles are resuspended. For information about preparing the reagents for use, refer to the system online help.

Preparing the DTT/Releasing Agent

Note Careful preparation of DTT/Releasing Agent is required to obtain accurate and consistent results since the absolute amount of DTT delivered to each test can affect results. The prepared DTT/Releasing Agent must be used within 108 hours after preparation.

1. Carefully transfer the contents of 1 vial of Releasing Agent into 1 vial of DTT. For convenience, the Releasing Agent can be poured or transferred by pipette into the DTT vial.
2. Firmly screw the cap on the DTT vial and invert the vial several times to mix.
3. Pour or pipette the entire contents of the DTT vial into the disposable ancillary reagent pack provided.
4. Place a pack seal on the disposable ancillary reagent pack. Ensure that the seal is centered over the openings of the pack, and press firmly on the adhesive portion of the seal.

Note DTT/Releasing Agent ReadyPack ancillary reagent packs are lot-number-specific. Do not use packs from one lot of DTT/Releasing Agent with any other lot of DTT/Releasing Agent.

Preparing the System

Ensure that the system has sufficient reagent packs loaded in the reagent compartment. The system automatically mixes reagent packs to maintain homogeneous suspension of the reagents. For information about loading reagent packs, refer to the system online help.

For automated dilutions, ensure that Atellica IM FoI DIL is loaded on the system.

Master Curve Definition

Before initiating calibration on each new lot of reagent, load the assay master curve and test definition values by scanning the MC TDEF 2D barcodes. For loading instructions, refer to the system online help.

Performing Calibration

For calibration of the Atellica IM FoI assay, use the calibrators provided with each kit.

Do not pour the calibrators back into the vials after calibration because evaporation could occur, which may affect performance.

Do not refill calibrator sample cups when the contents are depleted. If required, dispense fresh calibrators.

Calibration Frequency

Perform a calibration if one or more of the following conditions exist:

- When changing lot numbers of primary reagent packs.
- At the end of the lot calibration interval, for a specified lot of calibrated reagent on the system.
- At the end of the pack calibration interval, for calibrated reagent packs on the system.
- When indicated by quality control results.
- After major maintenance or service, if indicated by quality control results.

At the end of the onboard stability interval, replace the reagent pack on the system with a new reagent pack. Recalibration is not required, unless the lot calibration interval is exceeded.

Stability Interval	Days
Lot Calibration	14
Pack Calibration	4
Reagent Onboard Stability	7

For information about lot calibration and pack calibration intervals, refer to the system online help.

Follow government regulations or accreditation requirements for calibration frequency. Individual laboratory quality control programs and procedures may require more frequent calibration.

Preparing the Calibrators

Prepare calibrators using the following steps:

1. Add 3.0 mL of special reagent water into each vial using a precision pipet. Replace cap.

   Note For information about special reagent water requirements, refer to the system online help.

2. Let the vials stand for 15–20 minutes at room temperature to allow the lyophilized material to dissolve.
3. Gently mix and invert the vials to ensure homogeneity of the material.
4. For extended storage, aliquot and seal tightly. Store reconstituted material according to stability limits specified in *Storing the Specimen*. Do not store in a frost-free freezer.

   Note Before using frozen calibrators, allow the material to completely thaw. Gently mix and invert the vials to ensure homogeneity of the material. Use immediately and discard any remaining material.

Note Use calibrators within the stability limits specified in *Storing the Specimen* and discard any remaining material.

Calibration Procedure

The required sample volume for testing depends on several factors. For information about sample volume requirements, refer to the system online help.

Use the following lot-specific materials to perform calibration:

- For the master curve and assay test definitions, refer to the lot-specific master curve and test definition sheet MC TDEF provided with the assay reagents.
- Calibrators provided in an assay kit must only be used with reagents from that assay kit lot. Do not use calibrators from one assay kit with reagents from a different assay kit lot.
- For the calibrator definitions, refer to the lot-specific value sheet CAL LOT VAL provided with the calibrator materials.
- Generate lot-specific barcode labels to use with the calibrator samples.

For instructions about how to perform the calibration procedure, refer to the system online help.

Performing Quality Control

For quality control of the Atellica IM Fol assay, use an appropriate quality control material of known analyte concentration with a minimum of two levels (low and high) at least once during each day that samples are analyzed. For assistance in identifying a quality control material, refer to *Atellica® IM Quality Control Material Supplement* available on siemens-healthineers.com.

Additional quality control material can be used at the discretion of the laboratory. Use the quality control material in accordance with the quality control instructions for use.

In addition, perform quality control:

- Following a valid calibration
- With use of a new lot of reagent
- When troubleshooting test results that do not match clinical conditions or symptoms

Follow government regulations or accreditation requirements for quality control frequency. Individual laboratory quality control programs and procedures may require more frequent quality control testing.

Acceptable performance is achieved when the analyte values obtained are within the expected control interval for the system, as indicated by the manufacturer of the control material or within the interval determined by an internal laboratory quality control procedure.

Follow your laboratory's quality control procedures if the results obtained do not fall within the acceptable limits. For information about entering quality control definitions, refer to the system online help.

Taking Corrective Action

If the quality control results do not fall within the assigned values, do not report results. Perform corrective actions in accordance with established laboratory protocol. For suggested protocol, refer to the system online help.

Results

Calculation of Results

The system determines the result using the calculation scheme described in the system online help. The system reports results in ng/mL (common units) or nmol/L (SI units), depending on the units defined when setting up the assay.

Conversion formula: 1 ng/mL = 2.265 nmol/L

For information about results outside the specified measuring interval, refer to *Measuring Interval*.

Atellica IM Test Options

The Atellica IM FoI assay can be used for the following test options:

- FoI = system test generated whole blood folate concentration
- rFOL = predefined ratio test on the system for system-generated hemolysate folate value
- rbcFOL = predefined ratio test on the system for system-generated hemolysate RBC folate value (requires off-system hematocrit value)

  Note For information about creating the off-system hematocrit test, refer to the system online help.
- Manually calculated RBC Folate value using the rFOL predefined ratio value generated by the system for the hemolysate and the off-system hematocrit value
- Corrected Red Blood Cell Folate = system or manually calculated corrected RBC Folate

For information about entering the off-system and ratio test definitions, refer to the system online help.

Manual Calculation for Red Blood Cell Folate

Use this procedure to manually calculate RBC folate values using an off-system hematocrit value and the result from the rFOL test.

1. Prepare the RBC hemolysate as described in *Preparing the Red Blood Cell Hemolysate*.
2. Order the rFOL predefined ratio test.

   Note The rFOL predefined ratio test must use RBC hemolysate sample. Do not use other sample types.

3. Multiply the folate result for the hemolysate by 21 (a 1:21 dilution was made when preparing the RBC hemolysate). This value represents the folate concentration of whole blood in ng/mL.
4. Divide this result by the hematocrit, and multiply by 100 to adjust for the hematocrit, which is a percentage.

$$\text{RBC folate (ng/mL)} = \frac{\text{Folate result for hemolysate, (ng/mL)} \times 21}{\text{hematocrit}} \times 100$$

Example:
Hemolysate folate value = 5.7 ng/mL
Hematocrit = 43

$$\text{RBC folate (ng/mL)} = \frac{5.7 \times 21}{43} \times 100 = 278$$

Red Blood Cell Folate using the predefined system ratio test (rbcFOL)

1. Prepare the RBC hemolysate as described in *Preparing the Red Blood Cell Hemolysate*.
2. Order rbcFOL from the list of predefined ratio tests.

   Note The rbcFOL predefined ratio test must use RBC hemolysate sample. Do not use other sample types.
3. Manually enter the hematocrit value for the sample.
4. The system will display the calculated red blood cell folate value.

Corrected Red Blood Cell Folate

In most cases the serum folate values are very small compared to red blood cell folate values. However, occasionally serum folate values can be elevated. If the serum folate value is high and the red blood cell folate concentration is low, calculate the corrected RBC folate value according to the following equation:

$$\text{Corrected RBC folate (ng/mL)} = \underset{\text{(ng/mL)}}{\text{RBC folate}} - \underset{\text{(ng/mL)}}{\text{serum folate}} \left[\frac{(100 - \text{hematocrit})}{\text{hematocrit}}\right]$$

Example:
RBC folate = 210 ng/mL
Serum folate = 22 ng/mL
Hematocrit of the patient = 41

$$\text{Corrected RBC folate (ng/mL)} = 210 - 22 \left[\frac{(100 - 41)}{41}\right] = 210 - 32 = 178$$

For information about entering the off-system and ratio test definitions, refer to the system online help.

Dilutions

The assay measuring interval for serum is 0.35–24.00 ng/mL (0.79–54.36 nmol/L). For information about dilution options used to extend the reportable measuring interval up to 48.00 ng/mL (108.72 nmol/L), refer to the system online help.

Serum samples with folate levels > 24.00 ng/mL (> 54.36 nmol/L) must be diluted and retested to obtain accurate results.

Note Do not dilute the RBC hemolysate.

For automated dilutions, ensure that Atellica IM Fol DIL is loaded on the system. Ensure that sufficient sample volume is available to perform the dilution and that the appropriate dilution factor is selected when scheduling the test, as indicated in the table below.

For automatic dilutions, enter a dilution setpoint ≤ 24.00 ng/mL (≤ 54.36 nmol/L).

Sample	Dilution	Sample Volume (µL)
Serum	1:2	100

Interpretation of Results

Results of this assay should always be interpreted in conjunction with the patient's medical history, clinical presentation, and other findings.

Limitations

The following information pertains to limitations of the assay:

- Hemolysis significantly increases folate values in serum due to the high folate concentrations found in red blood cells.
- Methotrexate and leucovorin interfere with folate measurement because these drugs cross-react with folate binding proteins.

Expected Values

The reagent formulations used on the Atellica CI Analyzer are the same as those used on the ADVIA Centaur® system. Expected values were established using the ADVIA Centaur system and confirmed by assay comparison. Refer to *Assay Comparison*.

To determine the reference intervals for serum and RBC folate, data were obtained using 370 and 286 samples, respectively. For sample results in the indeterminate range [3.38–5.38 ng/mL (7.64–12.19 nmol/L)], clinical results and other diagnostic protocols should supplement folate results.

Category	N[a]	Median (ng/mL)	Range (ng/mL)	Median (nmol/L)	Range (nmol/L)
Serum folate					
Deficient[c]	65	1.54	0.35–3.37	3.49	0.79–7.63
Indeterminate[d]			3.38–5.38		7.64–12.19
Normal	305	12.51	> 5.38[b]	28.34	> 12.19
RBC folate					
Normal	286	425	280–791	963	634–1792

[a] Number of samples.
[b] Inner 97.5% of the distribution of apparently healthy individuals.
[c] Diagnosed by bone and/or peripheral blood smear pathology and other criteria including:
- megaloblastic anemia
- folate-deficient diet
- malabsorption
- alcoholism
- Tropical Sprue
- abnormal blood parameters including mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and hematocrit (HCT)

[d] Range between deficient and normal range.

Laboratories should consider these expected values as guidelines only. The data were obtained on apparently healthy males and females from the United States. Because of population demographic factors, diet, and assay methods, each laboratory should determine its own expected values for the diagnostic evaluation of patient results.[13]

Performance Characteristics

The reagents used on the Atellica CI Analyzer are the same as those used on the Atellica IM Analyzer. Some performance characteristics were established using the Atellica IM Analyzer.

Measuring Interval

The Atellica IM Fol assay is linear from 0.35–24.00 ng/mL (0.79–54.36 nmol/L). The lower limit of the measuring interval is defined by the analytical sensitivity. Report results below the measuring interval as < 0.35 ng/mL (< 0.79 nmol/L). When sample results exceed the measuring interval, refer to *Dilutions*.

Detection Capability

Analytical Sensitivity	0.35 ng/mL (0.79 nmol/L)
Limit of Blank (LoB)	0.40 ng/mL (0.91 nmol/L)

Limit of Detection (LoD)	0.70 ng/mL (1.59 nmol/L)
Limit of Quantitation (LoQ)	0.70 ng/mL (1.59 nmol/L)

Analytical sensitivity is defined as the concentration of folate that corresponds to the RLUs that are 2 standard deviations less than the mean RLUs of 20 replicate determinations of the folate zero standard. This response is an estimate of the minimum detectable concentration with 95% confidence.

The LoB corresponds to the highest measurement result likely to be observed for a blank sample with a probability of 95%.

The LoD corresponds to the lowest concentration of folate that can be detected with a probability of 95%.

The LoQ corresponds to the lowest amount of folate in a sample at which the within-laboratory CV is ≤ 20%.

Detection capability was determined in accordance with CLSI Document EP17-A2.[14]

Precision

Precision was determined in accordance with CLSI Document EP05-A3.[15] Samples were assayed on an Atellica CI Analyzer in duplicate in 2 runs per day for 20 days.

The following results are representative of the performance of the assay:

		Mean		Repeatability			Within-Laboratory Precision		
				SD[b]		CV[c]	SD		CV
Sample Type	N[a]	(ng/mL)	(nmol/L)	(ng/mL)	(nmol/L)	(%)	(ng/mL)	(nmol/L)	(%)
Serum A	80	1.02	2.31	0.057	0.129	5.6	0.073	0.165	7.2
Serum B	80	3.15	7.13	0.083	0.188	2.6	0.108	0.245	3.4
Serum C	80	5.28	11.96	0.165	0.374	3.1	0.220	0.498	4.2
Serum D	80	9.37	21.22	0.184	0.417	2.0	0.285	0.646	3.0
Serum E	80	16.49	37.35	0.463	1.049	2.8	0.735	1.665	4.5
Serum Control 1	80	2.60	5.89	0.070	0.159	2.7	0.123	0.279	4.7
Serum Control 2	80	7.11	16.10	0.190	0.430	2.7	0.301	0.682	4.2
Serum Control 3	80	11.68	26.46	0.277	0.627	2.4	0.524	1.187	4.5
Whole Blood Sample A	80	67.20	152.21	2.112	4.784	3.1	2.989	6.770	4.4
Whole Blood Sample B	80	106.99	242.33	2.370	5.368	2.2	3.973	8.999	3.7
Whole Blood Sample C	80	288.08	652.50	4.300	9.740	1.5	8.538	19.339	3.0
Whole Blood Sample D	80	433.54	981.97	7.660	17.350	1.8	10.175	23.046	2.3
Whole Blood Sample E	80	701.25	1588.33	4.617	10.458	0.7	14.149	32.047	2.0
Whole Blood Control 1	80	109.36	247.70	2.715	6.149	2.5	3.812	8.634	3.5

		Mean		Repeatability			Within-Laboratory Precision		
				SD[b]		CV[c]	SD		CV
Sample Type	N[a]	(ng/mL)	(nmol/L)	(ng/mL)	(nmol/L)	(%)	(ng/mL)	(nmol/L)	(%)
Whole Blood Control 2	80	298.31	675.67	4.509	10.213	1.5	6.725	15.232	2.3
Whole Blood Control 3	80	384.03	869.83	5.760	13.046	1.5	8.543	19.350	2.2

[a] Number of measurements.
[b] Standard deviation.
[c] Coefficient of variation.

Reproducibility

Reproducibility was determined in accordance with CLSI Document EP05-A3[15] using the Atellica CI Analyzer. Samples were assayed in replicates of 5 with 1 run per day for 5 days using 3 instruments and 3 reagent lots (225 measurements per sample).

The following results are representative of the performance of the assay:

		Repeatability		Between Day		Between Lot		Between Instrument		Reproducibility	
Sample	Mean ng/mL (nmol/L)	SD ng/mL (nmol/L)	CV (%)	SD ng/mL (nmol/L)	CV (%)	SD ng/mL (nmol/L)	CV (%)	SD ng/mL (nmol/L)	CV (%)	SD ng/mL (nmol/L)	CV (%)
Serum A	1.08 (2.45)	0.077 (0.174)	7.1	0.137 (0.310)	12.7	0.000 (0.000)	0.0	0.045 (0.102)	4.2	0.163 (0.369)	15.1
Serum B	3.36 (7.61)	0.131 (0.297)	3.9	0.119 (0.270)	3.5	0.031 (0.070)	0.9	0.115 (0.260)	3.4	0.213 (0.482)	6.3
Serum C	5.47 (12.39)	0.193 (0.437)	3.5	0.159 (0.360)	2.9	0.080 (0.181)	1.5	0.083 (0.188)	1.5	0.276 (0.625)	5.0
Serum D	9.75 (22.08)	0.354 (0.802)	3.6	0.215 (0.487)	2.2	0.167 (0.378)	1.7	0.193 (0.437)	2.0	0.486 (1.101)	5.0
Serum E	16.70 (37.83)	0.488 (1.105)	2.9	0.232 (0.525)	1.4	0.264 (0.598)	1.6	0.474 (1.074)	2.8	0.766 (1.735)	4.6
Serum Control 1	3.13 (7.09)	0.123 (0.279)	3.9	0.187 (0.424)	6.0	0.282 (0.639)	9.0	0.055 (0.125)	1.8	0.364 (0.824)	11.6
Serum Control 2	7.86 (17.80)	0.222 (0.503)	2.8	0.214 (0.485)	2.7	0.449 (1.017)	5.7	0.156 (0.353)	2.0	0.567 (1.284)	7.2
Serum Control 3	12.53 (28.38)	0.389 (0.881)	3.1	0.354 (0.802)	2.8	0.545 (1.234)	4.3	0.214 (0.485)	1.7	0.787 (1.783)	6.3

The assay was designed to have the following reproducibility:

Concentration Interval		Reproducibility
(ng/mL)	(nmol/L)	
3.00–20.00	6.80–45.30	≤ 20% CV

Assay results obtained at individual laboratories may vary from the data presented.

Collection Tube Comparison

Red Blood Cell Folate

Specimen equivalency was determined using weighted Deming linear regression in accordance with CLSI Document EP09-A3[16] using the Atellica IM Analyzer. The following results were obtained:

Specimen (y)	Reference Specimen (x)	Regression Equation	Sample Interval	N[a]
Dipotassium EDTA whole blood	Lithium heparin whole blood	y = 1.02x - 32.86 ng/mL (y = 1.02x - 74.43 nmol/L)	432.70–1103.69 ng/mL (980.07–2499.86 nmol/L)	90

[a] Number of samples tested.

Assay Comparison

Assay comparison was determined with the weighted Deming regression model in accordance with CLSI Document EP09c-ed3.[17]

Agreement of the assays may vary depending on the study design, comparative assay, and population tested.

Specimen	Comparative Assay (x)	Regression Equation	Sample Interval	N[a]	r[b]
Serum	Atellica IM Fol on Atellica IM Analyzer	y = 1.01x - 0.04 ng/mL (y = 1.01x - 0.09 nmol/L)	0.83–23.77 ng/mL (1.88–53.84 nmol/L)	115	0.995
Whole Blood	Atellica IM Fol on Atellica IM Analyzer	y = 0.96x + 15.28 ng/mL (y = 0.96x + 34.61 nmol/L)	61.87–676.35 ng/mL (140.14–1531.93 nmol/L)	102	0.989

[a] Number of samples tested.
[b] Correlation coefficient.

The assay is designed to have a correlation coefficient of ≥ 0.90 and a slope of 1.00 ± 0.10.

Interferences

The Atellica IM Fol assay is designed to have ≤ 10% interference for bilirubin and lipemia. Interfering substances at the levels indicated in the table below were tested in accordance with CLSI Document EP07-A2[18] using the Atellica IM Analyzer.

Bias is the difference in the results between the control sample (does not contain the interferent) and the test sample (contains the interferent) expressed in percent. Analyte results should not be corrected based on this bias.

Substance	Substance Test Concentration Common Units (SI Units)	Analyte Concentration ng/mL (nmol/L)	Bias (%)
Bilirubin, conjugated	20 mg/dL (341 µmol/L)	3.8 (8.61)	0.7
	20 mg/dL (341 µmol/L)	9.5 (21.52)	9.1
Bilirubin, unconjugated	20 mg/dL (341 µmol/L)	3.6 (8.15)	-5.3
	20 mg/dL (341 µmol/L)	9.8 (22.20)	-1.5
Lipemia (Intralipid®)	2000 mg/dL (22.7 mmol/L)	5.0 (11.33)	2.0
	2000 mg/dL (22.7 mmol/L)	8.8 (19.93)	3.0

Biotin was added to serum samples containing different concentrations of folate. These samples were tested against an appropriate control and the observed bias is presented in the following table:

Analyte Concentration ng/mL (nmol/L)	Biotin Test Level in Serum (ng/mL)								
	0	1	5	19	38	75	150	300	600
	% Bias								
9.23 (20.91)	—	1.4	-1.1	6.9	4.7	20.2	> MI[a]	> MI	> MI
17.37 (39.34)	—	1.3	1.3	3.6	8.3	35.3	> MI	> MI	> MI

[a] Measuring Interval

In addition, biotin was added to whole blood samples containing different concentrations of folate. These samples were tested against an appropriate control and the observed bias is presented in the following table:

Analyte Concentration ng/mL (nmol/L)	Biotin Test Level in Whole Blood (ng/mL)								
	0	1	5	19	38	75	150	300	600
	% Bias								
13.19 (29.88)	—	-6.8	-8.1	-8.5	-5.6	-3.4	-1.1	47.9	65.8
18.90 (42.81)	—	0.1	4.4	-0.2	4.3	7.6	12.4	> MI[a]	> MI

[a] Measuring Interval

Specimens that contain biotin at a concentration of 50 ng/mL (in serum) or 75 ng/mL (in whole blood) demonstrate a less than or equal to 10% change in results. Biotin concentrations greater than these may lead to falsely elevated results for patient samples.

The recommended adult daily dietary intake for biotin is 30 µg/day. Over the counter dietary supplements promoted for use in hair, skin, and nail health may contain 5–100 mg of biotin, with recommendations to take multiple pills per day. Pharmacokinetic studies in healthy adults have shown that, in subjects ingesting 5 mg, 10 mg, and 20 mg of biotin, serum concentrations of biotin can reach up to 73 ng/mL, 141 ng/mL, and 355 ng/mL, respectively.[19] Subjects who take up to 300 mg of biotin per day may have plasma biotin levels as high as 1160 ng/mL.[20]

Assay results obtained at individual laboratories may vary from the data presented.

Dilution Recovery

Serum samples were diluted onboard the Atellica CI Analyzer with Atellica IM Fol DIL. The following results are representative of the performance of the assay:

Sample	Dilution	Observed (ng/mL)	Expected (ng/mL)	Observed (nmol/L)	Expected (nmol/L)	Recovery (%)
1	1:2	34.16	39.82	77.37	90.19	85.8
2	1:2	35.08	42.19	79.46	95.56	83.1
3	1:2	40.66	43.48	92.09	98.48	93.5

Sample	Dilution	Observed (ng/mL)	Expected (ng/mL)	Observed (nmol/L)	Expected (nmol/L)	Recovery (%)
4	1:2	43.54	46.44	98.62	105.19	93.8
5	1:2	28.58	29.29	64.73	66.34	97.6

Assay results obtained at individual laboratories may vary from the data presented.

Spiking Recovery

Varying amounts of folate were added to 5 samples with endogenous folate levels of 1.05–1.35 ng/mL (2.38–3.06 nmol/L). The recoveries ranged from 87%–116% with a mean of 104%.

Sample	Amount Added (ng/mL)	Observed (ng/mL)	Amount Added (nmol/L)	Observed (nmol/L)	Recovery (%)
1	0.00	1.05	0.00	2.38	—
	3.57	4.65	8.09	10.53	108
	7.14	8.50	16.17	19.25	112
	10.70	12.20	24.24	27.63	115
	16.70	16.40	37.83	37.15	109
	19.00	18.30	43.04	41.45	108
	Mean				110
2	0.00	1.11	0.00	2.51	—
	3.57	4.65	8.09	10.53	105
	7.14	8.40	16.17	19.03	111
	10.70	12.30	24.24	27.86	116
	16.70	15.50	37.83	35.11	101
	19.00	19.60	43.04	44.39	116
	Mean				109
3	0.00	1.30	0.00	2.94	—
	3.57	4.45	8.09	10.08	95
	7.14	7.80	16.17	17.67	101
	10.70	11.40	24.24	25.82	106
	16.70	13.60	37.83	30.80	87
	19.00	17.10	43.04	38.73	99
	Mean				97
4	0.00	1.35	0.00	3.06	—
	2.98	3.97	6.75	8.99	94
	5.95	6.74	13.48	15.27	99

Sample	Amount Added (ng/mL)	Observed (ng/mL)	Amount Added (nmol/L)	Observed (nmol/L)	Recovery (%)
	8.90	9.40	20.16	21.29	99
	11.90	11.60	26.95	26.27	99
	14.90	14.50	33.75	32.84	103
	Mean				99
5	0.00	1.35	0.00	3.06	—
	2.98	4.11	6.75	9.31	101
	5.95	7.21	13.48	16.33	106
	8.90	10.10	20.16	22.88	108
	11.90	11.90	26.95	26.95	100
	14.90	14.90	33.75	33.75	105
	Mean				104
Mean					**104**

Results were established using the Atellica IM Analyzer. Assay results obtained at individual laboratories may vary from the data presented.

Standardization

The Atellica IM Fol assay is traceable to an internal standard manufactured using highly purified material (N-5-methyl tetrahydrofolate). Assigned values for calibrators are traceable to this standardization.

Technical Assistance

According to EU regulation 2017/746, any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority of the EU Member State in which the user and/or patient is established.

For customer support, contact your local technical support provider or distributor.

siemens-healthineers.com

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Definition of Symbols

The following symbols may appear on the product labeling:

Symbol	Symbol Title	Source	Symbol	Symbol Title	Source
	Manufacturer	5.1.1[a]		Authorized representative in the European Community	5.1.2[a]
	Use-by date	5.1.4[a]		Authorized representative in Switzerland	Proprietary
	Catalog number	5.1.6[a]		Batch code	5.1.5[a]
	Consult Instructions for Use	5.4.3[a]		Contains sufficient for <n> tests	5.5.5[a]
	Internet URL address to access the electronic instructions for use	Proprietary		Version of Instructions for Use	Proprietary
	*In vitro* diagnostic medical device	5.5.1[a]		Revision	Proprietary
RxOnly	Prescription device (US only)	FDA[b]		Unique Device Identifier	5.7.10[c]
	CE Marking with Notified Body	EU IVDR[d]		CE Marking	EU IVDR[d]
	Temperature limit	5.3.7[a]		Keep away from sunlight	5.3.2[a]
	Upper limit of temperature	5.3.6[a]		Lower limit of temperature	5.3.5[a]
	Do not re-use	5.4.2[a]		Do not freeze	Proprietary
	Recycle	1135[e]		This way up	0623[e]
	Biological risks	5.4.1[a]		Caution	5.4.4[a]
	Common Units	Proprietary		International System of Units	Proprietary
YYYY-MM-DD	Date format (year-month-day)	N/A	**YYYY-MM**	Date format (year-month)	N/A
	Document face up[f]	1952[e]		Handheld barcode scanner	Proprietary
	Target	Proprietary		Mixing of substances	5657[g]

Symbol	Symbol Title	Source	Symbol	Symbol Title	Source
CHECKSUM	Variable hexadecimal number that ensures the Master Curve and Calibrator definition values entered are valid.	Proprietary	←→	Interval	Proprietary
MATERIAL ID	Unique material identification number	Proprietary	MATERIAL	Material	Proprietary
CONTROL TYPE	Type of control	Proprietary	CONTROL NAME	Name of control	Proprietary
CONTROL LOT VAL	Quality control lot value	Proprietary	CAL LOT VAL	Calibrator lot value	Proprietary

[a] International Standard Organization (ISO). ISO 15223-1 Medical Devices- Symbols to be used with medical device labels, labelling and information to be supplied.
[b] Federal Register. Vol. 81, No 115. Wednesday, June 15, 2016. Rules and Regulations: 38911.
[c] ISO 15223-1:2020-04
[d] IVDR REGULATION (EU) 2017/746
[e] International Standard Organization (ISO). ISO 7000 Graphical symbols for use on equipment.
[f] Indicates Assay-eNote
[g] International Electrotechnical Commission (IEC). IEC 60417-1 Graphical symbols for use on equipment – Part 1: Overview and Application

Legal Information

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