TRAINING UPDATE

Lab Location:	SGMC & WAH	**Date Distributed:**	3/2/2017
Department:	Core Lab	**Due Date:**	3/28/2017
		Implementation:	**3/28/2017**

DESCRIPTION OF PROCEDURE REVISION

Name of procedure:

Lactate Dehydrogenase by Dimension Vista® System SGAH.C129 v2

Total Protein by Dimension Vista® System SGAH.C130 v2

Note: these have been converted to system SOPs

Description of change(s):

Both SOPs *(note QC change is already in effect)*

Section	Reason
Header	Add WAH
3.2	Specify anticoagulant, Remove specimen onboard stability
4,5,6	Remove individual section labeling instructions and add general one
6.1, 6.2	Update QC material and storage
6.4, 6.6	Replace LIS with Unity Real Time
7.2	Change freezer upper limit to -50C
10.5	Move patient review from section 6
15	Update to new standard wording
17	Update QC product

Total Protein SOP

Section	Reason
11.1	Add A/G ratio range

The revised SOPs will be implemented on March 28, 2017

Document your compliance with this training update by taking the quiz in the MTS system.

Technical SOP

Title	**Lactate Dehydrogenase by Dimension Vista® System**
Prepared by	Ashkan Chini Date: 7/12/2012
Owner	Robert SanLuis Date: 3/27/2014

Laboratory Approval	**Local Effective Date:**	
Print Name and Title	**Signature**	**Date**
Refer to the electronic signature page for approval and approval dates.		

Review		
Print Name	**Signature**	**Date**

Form revised 2/02/2007

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1. TEST INFORMATION

Assay	Method/Instrument	Local Code
Lactate Dehydrogenase, Serum / Plasma	Dimension Vista® System	LDH
Lactate Dehydrogenase, Body Fluid		FLD

Synonyms/Abbreviations
LD, LDH, LDI

Department
Chemistry

2. ANALYTICAL PRINCIPLE

The LDI method uses as a substrate L-lactate buffered at a pH of 9.4. Lactate dehydrogenase oxidizes the substrate in the presence of NAD+ to yield pyruvate and NADH which absorbs light at 340 nm. Lactate dehydrogenase activity is measured as a rate reaction at 340/700 nm, proportional to the amount of lactate dehydrogenase in the sample.

$$\text{L(+)-lactate} + \text{NAD+} \xrightarrow[\text{pH 9.4}]{\text{Lactate Dehydrogenase}} \text{Pyruvate} + H^+ + \text{NADH}$$

3. SPECIMEN REQUIREMENTS

3.1 Patient Preparation

Component	Special Notations
Fasting/Special Diets	N/A
Specimen Collection and/or Timing	Normal procedures for collecting and storing serum, plasma and body fluid may be used for samples to be analyzed by this method.
Special Collection Procedures	N/A
Other	N/A

3.2 Specimen Type & Handling

Criteria		
Type -Preferred **-Other Acceptable**	Plasma (Lithium Heparin), Body Fluid Serum	
Collection Container	Plasma: Mint green top tube (PST) Serum: Red top tube, Serum separator tube (SST) Body Fluid: Sterile/Clean container or tube	
Volume - Optimum **- Minimum**	1.0 mL 0.5 mL	
Transport Container and Temperature	Collection container or Plastic vial at room temperature	
Stability & Storage Requirements	Room Temperature:	3 days
	Refrigerated:	Not recommended
	Frozen:	Not recommended
Timing Considerations	Serum or plasma should be physically separated from cells as soon as possible with a maximum limit of two hours from the time of collection.	

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Criteria	
Unacceptable Specimens & Actions to Take	Specimens that are unlabeled, improperly labeled, or those that do not meet the stated criteria are unacceptable. Request a recollection and credit the test with the appropriate LIS English text code for "test not performed" message. Examples: Quantity not sufficient-QNS; Wrong collection-UNAC. Document the request for recollection in the LIS.
Compromising Physical Characteristics	Reject hemolyzed samples and request a recollection. Credit the test with the appropriate LIS English text code explanation of HMT (Specimen markedly hemolyzed)
Other Considerations	Allow Red Top or SST to clot completely prior to centrifugation.

NOTE: Labeling requirements for all reagents, calibrators and controls include: (1) Open date, (2) Substance name, (3) Lot number, (4) Date of preparation, (5) Expiration date, (6) Initials of tech, and (7) Any special storage instructions. Check all for visible signs of degradation. When placed onboard the analyzer, the instrument captures the date / time loaded and calculates and tracks the opened expiration.

4. REAGENTS

The package insert for a new lot of kits must be reviewed for any changes before the kit is used. A current Package Insert is included as a Related Document.

4.1 Reagent Summary

Reagents	Supplier & Catalog Number
Lactate Dehydrogenase	Siemens, Flex® reagent cartridge, Cat. No. K2054
Enzyme Diluent	Siemens Diagnostics Healthcare REF: 790035901

4.2 Reagent Preparation and Storage

Reagent	**Lactate Dehydrogenase**
Container	Reagent cartridge
Storage	Store at 2-8°C
Stability	• Reagent is stable until expiration date stamped on the reagent cartridges. • Sealed wells on the instrument are stable for 30 days. • Once wells 1 - 8 have been entered by the instrument, they are stable for 3 days. • Once wells 9 - 12 have been entered by the instrument, they are stable for 6 days.
Preparation	All reagents are liquid and ready to use.

Reagent	**Enzyme Diluent**
Container	Reagent vial
Storage	Store at 2-8°C
Stability	• Stable until expiration date stamped on the reagent vial. • Discard after 7 days following reconstitution or immediately if visible turbidity appears.
Preparation	• Remove vial from refrigerator and proceed with next step. • Remove stopper and volumetrically add 10.0 mL of reagent grade water. • Replace stopper and invert gently 10 times. • Sit vials for 15 minutes, then invert gently 10 times. • Sit vials for an additional 15 minutes, then invert 10 times and swirl gently. • Use immediately or store at 2-8°C. Before use, allow to come to room temperature, then invert 10 times and swirl gently.

5. CALIBRATORS/STANDARDS

5.1 Calibrators/Standards Used

Calibrator	Supplier and Catalog Number
ENZ 5 CAL	Siemens Dimension Vista®, Cat. No. KC350

5.2 Calibrator Preparation and Storage

Calibrator	ENZ 5 CAL
Preparation	Calibrator is ready for use. No preparation is required.
Storage/Stability	• Store at 2-8°C • **Unopened calibrator** is stable until expiration date stamped on the box. • **Opened Calibrator:** once the stopper of the vial is punctured, assigned values are stable for 7 days when stored on board the Dimension Vista System.

5.3 Calibration Parameter

Criteria	**Special Notations**
Reference Material	ENZ 5 CAL
Assay Range	6 – 1000 U/L

Suggested Calibration Level	See Reagent Package Insert for lot specific assigned values in U/L
Frequency	• Every new reagent cartridge lot. • Every 90 days for any one lot • When major maintenance is performed on the analyzer. • When control data indicates a significant shift in assay.
Calibration Scheme	2 levels, n = 5

5.4 Calibration Procedure

Auto Calibration:

1. Place the required calibrator vials in a carrier. Make sure the barcode labels are entirely visible through the slots.
2. Place the carrier in the loading area.
3. Position the carrier with the labels facing away from the user.
4. Press the **Load** button.
5. Automatic calibration requires that calibrators be on the instrument. As the time for processing approaches, the instrument reviews onboard inventory for the appropriate calibrators.

Manual Calibration:

1. Verify that calibrators and reagents are in inventory on the instrument.
2. Press **System > Method Summary > Calibration**.
3. Select a method from the sidebar menu. Press the **Order Calibration** button on the screen.
4. Verify that the information on the screen is correct. Verify that the calibrator lot is correct using the drop-down menu.
   a. When calibrating using Vials press **OK**.
   b. When calibrating using Cups, check the Use Cups box. This displays the rack and cup position fields. For additional cups use the positions in ascending order. Be sure to use the number of calibration levels and cups as specified in the method IFU. Scan the rack barcode and place calibrator cups in an adapter in position 1 on a rack. Press **OK** and load the rack on the instrument.
5. The status field in the calibration screen changes sequentially to Awaiting Scheduling, Preparing Calibrators and Processing.

5.5 Tolerance Limits

IF……	THEN……
If result fall within assay-specific specification, and QC values are within acceptable limits,	proceed with analysis
If result falls outside assay-specific specification, or QC values are out of Acceptable limits,	troubleshoot the assay and/or instrument and repeat calibration

6. QUALITY CONTROL

6.1 Controls Used

Controls	Supplier and Catalog Number
Liquid Assayed Multiqual® Levels 1 and 3	Bio-Rad Laboratories Cat. No. 337 and 339

6.2 Control Preparation and Storage

Control	Liquid Assayed Multiqual® Levels 1 and 3
Preparation	Allow the frozen control to stand at room temperature (18-25°C) for 30 minutes or until completely thawed. Swirl the contents gently to ensure homogeneity. (Do not use a mechanical mixer) Use immediately. After each use, promptly replace the stopper and return to 2-8°C storage.
Storage/Stability	Frozen controls are stable until the expiration date at -20 to -50°C. Thawed and Unopened: When this product is stored at 2-8°C and the stopper is not punctured, it will be stable for 30 days for total protein This product can be used for 7 days when stored on-board the Siemens Dimension Vista at 2- 8°C. Thawed and Opened: Once the product stopper is punctured, all analytes will be stable for 5 days when stored at 2- 8°C. Store away from light.

6.3 Frequency

Analyze all levels of QC material after every calibration and each day of testing (notated on the QC frequency sheets posted on the instruments).

Refer to the Dimension Vista® QC Schedule in the Laboratory policy Quality Control Program and in the Dimension Vista® Quick Reference Guide.

6.4 Tolerance Limits and Criteria for Acceptable QC

Step	Action
1	Acceptable ranges for QC are programmed into the instrument's Quality Control software system and Unity Real Time, and may be posted near the instrument for use during computer downtime.

Step	Action
2	**Run Rejection Criteria** • Anytime the established parameters are exceeded (if one QC result exceeds 2 SD), the run is considered out of control (failed) and patient results must not be reported. • The technologist must follow the procedure in the Laboratory QC Program to resolve the problem.
3	**Corrective Action**: • All rejected runs must be effectively addressed through corrective action. Steps taken in response to QC failures must be documented. Patient samples in failed analytical runs must be reanalyzed according to the Laboratory QC Program. Supervisors may override rejection of partial or complete runs only with detailed documentation and criteria for overrides that are approved by the Medical Director. Consult corrective action guidelines in Laboratory QC Program. Follow corrective action guidelines in the Laboratory QC Program. • Corrective action documentation must follow the Laboratory Quality Control Program.
4	**Review of QC** • QC must be reviewed weekly by the Group Lead or designee and monthly by the Supervisor/Manager or designee. • If the SD and/or CV are greater than established ranges, investigate the cause for the imprecision and document implementation of corrective actions.

6.5 Documentation

- QC tolerance limits are programmed into the instrument and Unity Real Time; it calculates cumulative mean, SD and CV and stores all information for easy retrieval.Quality control records are reviewed daily at the bench, weekly by the Group Lead or designee, and monthly by the Supervisor/Manager or designee.
- Refer to complete policies and procedures for QC documentation and for record retention requirements in the Laboratory QC Program.

6.6 Quality Assurance Program

- Each new lot number of reagent or new shipment of the same lot of reagent must be tested with external control materials and previously analyzed samples. Performance of the new lot must be equivalent to the previous lot; utilize published TEA for acceptability criteria.

- Training must be successfully completed and documented prior to performing this test. This procedure must be incorporated into the departmental competency assessment program.
- The laboratory participates in CAP proficiency testing. All proficiency testing materials must be treated in the same manner as patient samples.
- Monthly QC must be presented to the Medical Director or designee for review and signature.
- Monthly QC mean and SD are sent to Bio-Rad Laboratories for peer group comparison.
- Consult the Laboratory QC Program for complete details.

7. EQUIPMENT and SUPPLIES

7.1 Assay Platform

Dimension Vista® System

7.2 Equipment

- Refrigerator capable of sustaining 2–8°C.
- Freezer capable of sustaining range not to exceed -20 to -50°C.
- Centrifuge

7.3 Supplies

- Aliquot Plates
- System Fluids
- Assorted calibrated pipettes (MLA or equivalent) and disposable tips

8. PROCEDURE

LDI Flex® reagent cartridge Cat. No. K2054 is required to perform this test.

Lactate Dehydrogenase is performed on the Dimension Vista® System after the method is calibrated (see Reference Material in Calibration section) and Quality Controls are acceptable.

NOTE: For all procedures involving specimens, buttoned lab coats, gloves, and face protection are required minimum personal protective equipment. Report all accidents to your supervisor.

8.1	Sample Processing
1.	A sample rack holding tubes or cups is placed on the rack input lane.

8.1	Sample Processing
2.	The sample shuttle moves the rack to the barcode reader which identifies the rack and samples to the system.
3.	The rack moves into the sample server and to the rack positioner.
4.	At the same time, aliquot plates move from the aliquot loader into position.
5.	The aliquot probe aspirates the sample from the tubes or cups and dispenses it into the wells of the aliquot plates.
6.	After each aspirate-dispense action, the probe is thoroughly rinsed inside and out to prevent sample carryover.
7.	When sample aspiration is completed, the sample server moves the rack back to the sample shuttle, where it is placed on the output lane and can be removed by the operator.

8.2	Specimen Testing
1.	For QC placement and frequency, refer to the Dimension Vista® QC Schedule in the Laboratory QC Program.
2.	Follow the instructions, outlined in the Dimension Vista® Operator's Manual
3.	The instrument reporting system contains error messages to warn the user of specific malfunctions. Results followed by such error messages should be held for follow-up. Refer to the Dimension Vista® system manual "Error messages" section for troubleshooting.
4.	Follow protocol in Section 10.5 "Repeat criteria and resulting" for samples with results above or below the Analytical Measurement Range (AMR). Investigate any failed delta result and repeat, if necessary.
5.	Append the appropriate English text code qualifier messages to any samples requiring a comment regarding sample quality and/or any other pertinent factors.

Test Conditions	
Sample Volume:	4.1 µL
Reagent 1 Volume:	53.7 µL
Reagent 2 Volume:	25.3 µL
Reaction Time:	7.2 minutes
Test Temperature:	37° C
Wavelength:	340 & 700 nm
Type of measurement:	Bichromatic rate

9. CALCULATIONS

The instrument automatically calculates the concentration of Lactate Dehydrogenase in U/L.

10. REPORTING RESULTS AND REPEAT CRITERIA

10.1 Interpretation of Data

None required

10.2 Rounding

No rounding is necessary. Instrument reports results as a whole number.

10.3 Units of Measure

U/L

10.4 Clinically Reportable Range (CRR)

6 – 20,000 U/L

10.5 Review Patient Data

Each result is reviewed for error messages. Refer to the Dimension Vista system manual "Error messages" section for troubleshooting. Resolve any problems noted before issuing patient reports.

10.6 Repeat Criteria and Resulting

All repeats must replicate the original result within the total allowable error (TEa) of the assay. Refer to TEa listing for specific information.

Values that fall within the AMR or CRR may be reported without repeat. Values that fall outside these ranges must be repeated.

IF the result is …	THEN…
< 6 U/L	Assure there is sufficient sample devoid of bubbles, cellular debris, and/or fibrin clots. Report as: < 6 U/L
≥ 1,000 U/L	**On Board Automated Dilution:** Results ≥ 1,000 U/L will automatically have repeat testing performed into the instrument using dilution factor of 4. No multiplication is necessary.

> 4,000 U/L	**Manual Dilution**: Using the primary tube, make the smallest dilution possible to bring the raw data within the AMR. Maximum allowable dilution: x 20 **DILUENT**: Enzyme diluent Enter dilution factor as a whole number. Re-assay. Readout is corrected for dilution.
> 20,000 U/L	If the recommended dilution does not give results within the clinically reportable range, report as: "> 20,000 U/L-REP" Bring to the attention of your supervisor prior to releasing result.

Message	Code
Verified by repeat analysis	Append –REP to the result.

11. EXPECTED VALUES

11.1 Reference Ranges

Serum / Plasma:
Male: 87 – 241 U/L
Female: 84 – 246 U/L

Body Fluid: Peritoneal Fluid < 63 U/L

Pleural Fluid: Exudates > 113 U/L
Transudates < 113 U/L

11.2 Critical Values

None established

11.3 Standard Required Messages

None established

12. CLINICAL SIGNIFICANCE

Lactate dehydrogenase (LD) is present in the cytoplasm of all cells in the body. The concentration of LD in tissues is several hundred-fold higher than in serum or plasma and even a small amount of tissue damage can lead to an elevation in LD activity. This makes LD especially useful in the diagnosis and monitoring of disease states where tissue turnover is accelerated such as the liver, cardiac muscle, skeletal muscle, kidneys, and erythrocytes.

LD is elevated in myocardial or pulmonary infarction, leukemias, hemolytic anemias, non-viral hepatitis, sickle cell disease, lymphoma, renal infarction, acute pancreatitis and any condition that results in the leaking of cytoplasm. It is moderately elevated in cirrhosis, obstructive jaundice, renal disease, skeletal muscle diseases, neoplastic diseases and congestive heart failure. LD is markedly elevated in megaloblastic and pernicious anemia, metastatic carcinoma, viral hepatitis, shock, hypoxia and extreme hyperthermia.

13. PROCEDURE NOTES

- **FDA Status:** FDA Approved/cleared
- **Validated Test Modifications:** None

The instrument reporting system contains error messages to warn the operator of specific malfunctions. Any report slip containing such error messages should be held for follow-up. Refer to your Dimension Vista Operator's Guide.

The expected maximum observed standard deviations for repeatability using n = 5 replicates at the following Lactate Dehydrogenase concentrations are:

LDI Concentration	Acceptable S.D. Maximum
121 U/L	>11 U/L
401 U/L	>32 U/L

14. LIMITATIONS OF METHOD

14.1 Analytical Measurement Range (AMR)

6 – 1000 U/L

14.2 Precision

Material	Mean U/L	Standard Deviation (%CV)	
		Repeatability	Within-Lab
BioRad Multiqual			
Level 1	111	2.5 (2.3)	3.0 (2.8)
Level 2	166	3.2 (2.0)	4.1 (2.5)
Level 3	388	7.4 (1.9)	9.0 (2.4)

14.3 Interfering Substances

Hemoglobin (hemolysate) at 50 mg/dL increases LDI results by 16% at a lactate dehydrogenase activity concentration of 300 U/L and 500 U/L.
Dopamine at 65 mg/dL increases LDI results by 113% at a lactate dehydrogenase activity of 300 U/L.

HIL Interference:
The LDI method was evaluated for interference according to CLSI/NCCLS EP7-A2. Bias, defined as the difference between the control sample (does not contain interferent) and the test sample (contains interferent), is shown in the table below. Bias exceeding 10% is considered "interference".

Substance tested	Substance Concentration	LDI U/L	Bias %
Hemoglobin (hemolysate)	50 mg/dL	300, 500	16
Bilirubin (unconjugated)	80 mg/dL	300, 500	<10
Bilirubin (conjugated)	80 mg/dL	300, 500	<10
Lipemia Intralipid®	1000 mg/dL 3000 mg/dL	300, 500	<10

14.4 Clinical Sensitivity/Specificity/Predictive Values

Not available

15. SAFETY

Refer to your local and corporate safety manuals and Safety Data Sheet (SDS) for detailed information on safety practices and procedures and a complete description of hazards.

16. RELATED DOCUMENTS

1. Dimension Vista® Clinical Chemistry System Operator's Manual
2. Dimension Vista® Calibration/Verification Procedure
3. Dimension Vista® Cal Accept Guidelines
4. Dimension Vista® Calibration summary
5. Dimension Vista® Sample Processing, Startup and Maintenance procedure
6. Laboratory Quality Control Program
7. QC Schedule for Siemens Dimension Vista®
8. Laboratory Safety Manual
9. Safety Data Sheets (SDS)
10. Dimension Vista® Limits Chart (AG.F200)
11. Quest Diagnostics Records Management Procedure
12. Dimension Vista® System Error Messages Chart
13. Centrifuge Use, Maintenance and Functions Checks (Lab policy)
14. Hemolysis, Icteria and Lipemia Interference (Lab policy)
15. Repeat Testing Requirement (Lab policy)
16. Current Allowable Total Error Specifications
    at http://questnet1.qdx.com/Business_Groups/Medical/qc/docs/qc_bpt_tea.xls
17. Current package insert LDI Flex® Reagent Cartridge K2054

17. REFERENCES

1. Package Insert, LDI Flex® Reagent Cartridge K2054, Siemens Healthcare Diagnostics Inc., 8/20/2013.
2. Package Insert, ENZ 5 CAL, Siemens Healthcare Diagnostics Inc., 03/2009.
3. Package Insert, Liquid Assayed Multiqual® Chemistry Controls, Bio-Rad Laboratories, 09/2015.
4. Package Insert, Enzyme Diluent, Siemens Healthcare Diagnostics Inc., 10/2012.
5. Quest Diagnostics SOP ID 300SA355, Lactate Dehydrogenase

18. REVISION HISTORY

Version	Date	Section	Reason	Reviser	Approval
000	3/27/14		Update owner	L Barrett	R SanLuis
000	3/27/14	5.2	Remove 30 day stability	A Chini	R SanLuis
000	3/27/14	16	Update titles	L Barrett	R SanLuis
000	3/27/14	Footer	Version # leading zero's dropped due to new EDCS in use as of 10/7/13.	L Barrett	R SanLuis
1	2/13/17	Header	Add WAH	L Barrett	R SanLuis
1	2/13/17	3.2	Specify anticoagulant, remove specimen onboard stability	L Barrett	R SanLuis
1	2/13/17	4,5,6	Remove individual section labeling instructions and add general one	L Barrett	R SanLuis
1	2/13/17	6.1, 6.2	Update QC material and storage	L Barrett	R SanLuis
1	2/13/17	6.4, 6.6	Replace LIS with Unity Real Time	L Barrett	R SanLuis
1	2/13/17	7.2	Change freezer upper limit to -50C	L Barrett	R SanLuis
1	2/13/17	10.5	Move patient review from section 6	L Barrett	R SanLuis
1	2/13/17	15	Update to new standard wording	L Barrett	R SanLuis
1	2/13/17	17	Update QC product and PI dates	L Barrett	R SanLuis

19. ADDENDA

None

Technical SOP

Title	**Total Protein by Dimension Vista® System**
Prepared by	Ashkan Chini Date: 7/12/2012
Owner	Robert SanLuis Date: 3/27/2014

Laboratory Approval	**Local Effective Date:**	
Print Name and Title	**Signature**	**Date**
Refer to the electronic signature page for approval and approval dates.		

Review		
Print Name	**Signature**	**Date**

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1. TEST INFORMATION

Assay	Method/Instrument	Local Code
Total Protein, Serum / Plasma	Dimension Vista® System	TP
Total Protein, Body Fluid		FTP

Synonyms/Abbreviations
TP, included in Batteries/Packages: COMP, LIVP

Department
Chemistry

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2. ANALYTICAL PRINCIPLE

The total protein method is a modification of the biuret reaction first introduced by Kingsley and later modified by Henry and presented as the method of choice for serum by Henry. This method incorporates tartrate as a complexing agent to prevent precipitation of Cu(OH)2. Serum blanking increases method sensitivity and minimizes spectral interference from lipemia. Cupric ion (Cu++) reacts with the peptide linkages of protein in a basic solution. The blue copper (II) protein complex thus formed is proportional to the total protein concentration in the sample and is measured using a bichromatic (540, 700 nm) endpoint technique.

$$Cu^{++} + \text{Protein} \xrightarrow{OH^-} \text{complex}$$

(absorbs at 540 nm)

3. SPECIMEN REQUIREMENTS

3.1 Patient Preparation

Component	Special Notations
Fasting/Special Diets	N/A
Specimen Collection and/or Timing	Normal procedures for collecting and storing serum, plasma and body fluid may be used for samples to be analyzed by this method.
Special Collection Procedures	N/A
Other	N/A

3.2 Specimen Type & Handling

Criteria		
Type -Preferred **-Other Acceptable**	Plasma (Lithium Heparin), Body Fluid Serum	
Collection Container	Plasma: Mint green top tube (PST) Serum: Red top tube, Serum separator tube (SST) Body Fluid: Sterile/Clean container or tube	
Volume - Optimum **- Minimum**	1.0 mL 0.5 mL	
Transport Container and Temperature	Collection container or Plastic vial at room temperature	
Stability & Storage Requirements	Room Temperature:	8 hours
	Refrigerated:	72 hours
	Frozen:	6 months

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Criteria	
Timing Considerations	Serum or plasma should be physically separated from cells as soon as possible with a maximum limit of two hours from the time of collection.
Unacceptable Specimens & Actions to Take	Specimens that are unlabeled, improperly labeled, or those that do not meet the stated criteria are unacceptable. Request a recollection and credit the test with the appropriate LIS English text code for "test not performed" message. Examples: Quantity not sufficient-QNS; Wrong collection-UNAC. Document the request for recollection in the LIS.
Compromising Physical Characteristics	Gross hemolysis. Reject sample and request a recollection. Credit the test with the appropriate LIS English text code explanation of HMT (Specimen markedly hemolyzed)
Other Considerations	Allow Red Top or SST to clot completely prior to centrifugation.

NOTE: Labeling requirements for all reagents, calibrators and controls include: (1) Open date, (2) Substance name, (3) Lot number, (4) Date of preparation, (5) Expiration date, (6) Initials of tech, and (7) Any special storage instructions. Check all for visible signs of degradation. When placed onboard the analyzer, the instrument captures the date / time loaded and calculates and tracks the opened expiration.

4. REAGENTS

The package insert for a new lot of kits must be reviewed for any changes before the kit is used. A current Package Insert is included as a Related Document.

4.1 Reagent Summary

Reagents	Supplier & Catalog Number
Total Protein	Siemens, Flex® reagent cartridge, Cat. No. K1073

4.2 Reagent Preparation and Storage

Reagent	**Total Protein**
Container	Reagent cartridge
Storage	Store at 2-8° C
Stability	• Reagent is stable until expiration date stamped on the reagent cartridges. • Sealed wells on the instrument are stable for 30 days. • Once wells 1 - 12 have been entered by the instrument, they are stable for 7 days.
Preparation	All reagents are liquid and ready to use.

5. CALIBRATORS/STANDARDS

5.1 Calibrators/Standards Used

Calibrator	Supplier and Catalog Number
CHEM 4 CAL	Siemens Dimension Vista®, Cat. No. KC140

5.2 Calibrator Preparation and Storage

Calibrator	CHEM 4 CAL
Preparation	CHEM 4 CAL is ready for use. No preparation required.
Storage/Stability	• Store at 2-8° C • **Unopened calibrator** is stable until expiration date stamped on the box. • **Opened Calibrator:** once the stopper of the vial is punctured, assigned values are stable for 7 days when stored on board the Dimension Vista System.

5.3 Calibration Parameter

Criteria	Special Notations
Reference Material	CHEM 4 CAL
Assay Range	0.0 – 12.0 g/dL
Suggested Calibration Level	See Reagent Package Insert for lot specific assigned values in g/dL
Frequency	• Every new reagent cartridge lot. • Every 90 days for any one lot • When major maintenance is performed on the analyzer. • When control data indicates a significant shift in assay.
Calibration Scheme	2 levels, n = 3

5.4 Calibration Procedure

Auto Calibration:

1. Place the required calibrator vials in a carrier. Make sure the barcode labels are entirely visible through the slots.
2. Place the carrier in the loading area.
3. Position the carrier with the labels facing away from the user.
4. Press the **Load** button.
5. Automatic calibration requires that calibrators be on the instrument. As the time for processing approaches, the instrument reviews onboard inventory for the appropriate calibrators.

Manual Calibration:

1. Verify that calibrators and reagents are in inventory on the instrument.
2. Press **System > Method Summary > Calibration**.
3. Select a method from the sidebar menu. Press the **Order Calibration** button on the screen.
4. Verify that the information on the screen is correct. Verify that the calibrator lot is correct using the drop-down menu.
   a. When calibrating using Vials press **OK**.
   b. When calibrating using Cups, check the Use Cups box. This displays the rack and cup position fields. For additional cups use the positions in ascending order. Be sure to use the number of calibration levels and cups as specified in the method IFU. Scan the rack barcode and place calibrator cups in an adapter in position 1 on a rack. Press **OK** and load the rack on the instrument.
5. The status field in the calibration screen changes sequentially to Awaiting Scheduling, Preparing Calibrators and Processing.

5.5 Tolerance Limits

IF……	THEN……
If result fall within assay-specific specification, and QC values are within acceptable limits,	proceed with analysis
If result falls outside assay-specific specification, or QC values are out of Acceptable limits,	troubleshoot the assay and/or instrument and repeat calibration

6. QUALITY CONTROL

6.1 Controls Used

Controls	Supplier and Catalog Number
Liquid Assayed Multiqual® Levels 1 and 3	Bio-Rad Laboratories Cat. No. 337 and 339

6.2 Control Preparation and Storage

Control	Liquid Assayed Multiqual® Levels 1 and 3
Preparation	Allow the frozen control to stand at room temperature (18-25°C) for 30 minutes or until completely thawed. Swirl the contents gently to ensure homogeneity. (Do not use a mechanical mixer) Use immediately. After each use, promptly replace the stopper and return to 2-8°C storage.

Storage/Stability	Frozen controls are stable until the expiration date at -20 to -50°C. Thawed and Unopened: When this product is stored at 2-8°C and the stopper is not punctured, it will be stable for 30 days for total protein This product can be used for 7 days when stored on-board the Siemens Dimension Vista at 2- 8°C. Thawed and Opened: Once the product stopper is punctured, all analytes will be stable for 5 days when stored at 2- 8°C. Store away from light.

6.3 Frequency

Analyze all levels of QC material after every calibration and each day of testing (notated on the QC frequency sheets posted on the instruments).

Refer to the Dimension Vista® QC Schedule in the Laboratory policy Quality Control Program and in the Dimension Vista® Quick Reference Guide.

6.4 Tolerance Limits and Criteria for Acceptable QC

Step	Action
1	Acceptable ranges for QC are programmed into the instrument's Quality Control software system and Unity Real Time, and may be posted near the instrument for use during computer downtime.
2	**Run Rejection Criteria** • Anytime the established parameters are exceeded (if one QC result exceeds 2 SD), the run is considered out of control (failed) and patient results must not be reported. • The technologist must follow the procedure in the Laboratory QC Program to resolve the problem.
3	**Corrective Action**: • All rejected runs must be effectively addressed through corrective action. Steps taken in response to QC failures must be documented. Patient samples in failed analytical runs must be reanalyzed according to the Laboratory QC Program. Supervisors may override rejection of partial or complete runs only with detailed documentation and criteria for overrides that are approved by the Medical Director. Consult corrective action guidelines in Laboratory QC Program. Follow corrective action guidelines in the Laboratory QC Program. • Corrective action documentation must follow the Laboratory Quality Control Program.

Step	Action
4	**Review of QC** • QC must be reviewed weekly by the Group Lead or designee and monthly by the Supervisor/Manager or designee. • If the SD and/or CV are greater than established ranges, investigate the cause for the imprecision and document implementation of corrective actions.

6.5 Documentation

- QC tolerance limits are programmed into the instrument and Unity Real Time; it calculates cumulative mean, SD and CV and stores all information for easy retrieval.
- Quality control records are reviewed daily at the bench, weekly by the Group Lead or designee, and monthly by the Supervisor/Manager or designee.
- Refer to complete policies and procedures for QC documentation and for record retention requirements in the Laboratory QC Program.

6.6 Quality Assurance Program

- Each new lot number of reagent or new shipment of the same lot of reagent must be tested with external control materials and previously analyzed samples. Performance of the new lot must be equivalent to the previous lot; utilize published TEA for acceptability criteria.
- Training must be successfully completed and documented prior to performing this test. This procedure must be incorporated into the departmental competency assessment program.
- The laboratory participates in CAP proficiency testing. All proficiency testing materials must be treated in the same manner as patient samples.
- Monthly QC must be presented to the Medical Director or designee for review and signature.
- Monthly QC mean and SD are sent to Bio-Rad Laboratories for peer group comparison.
- Consult the Laboratory QC Program for complete details.

7. EQUIPMENT and SUPPLIES

7.1 Assay Platform

Dimension Vista® System

7.2 Equipment

- Refrigerator capable of sustaining 2–8°C.
- Freezer capable of sustaining range not to exceed -20 to -50°C.
- Centrifuge

7.3 Supplies

- Aliquot Plates
- System Fluids
- Assorted calibrated pipettes (MLA or equivalent) and disposable tips

8. PROCEDURE

TP Flex® reagent cartridge Cat. No. K1073 is required to perform this test.

Total Protein is performed on the Dimension Vista® System after the method is calibrated (see Reference Material in Calibration section) and Quality Controls are acceptable.

NOTE: For all procedures involving specimens, buttoned lab coats, gloves, and face protection are required minimum personal protective equipment. Report all accidents to your supervisor.

8.1	Sample Processing
1.	A sample rack holding tubes or cups is placed on the rack input lane.
2.	The sample shuttle moves the rack to the barcode reader which identifies the rack and samples to the system.
3.	The rack moves into the sample server and to the rack positioner.
4.	At the same time, aliquot plates move from the aliquot loader into position.
5.	The aliquot probe aspirates the sample from the tubes or cups and dispenses it into the wells of the aliquot plates.
6.	After each aspirate-dispense action, the probe is thoroughly rinsed inside and out to prevent sample carryover.
7.	When sample aspiration is completed, the sample server moves the rack back to the sample shuttle, where it is placed on the output lane and can be removed by the operator.

8.2	Specimen Testing
1.	For QC placement and frequency, refer to the Dimension Vista® QC Schedule in the Laboratory QC Program.
2.	Follow the instructions, outlined in the Dimension Vista® Operator's Manual
3.	The instrument reporting system contains error messages to warn the user of specific malfunctions. Results followed by such error messages should be held for follow-up. Refer to the Dimension Vista® system manual "Error messages" section for troubleshooting.

8.2	Specimen Testing
4.	Follow protocol in Section 10.5 "Repeat criteria and resulting" for samples with results above or below the Analytical Measurement Range (AMR). Investigate any failed delta result and repeat, if necessary.
5.	Append the appropriate English text code qualifier messages to any samples requiring a comment regarding sample quality and/or any other pertinent factors.

Test Conditions	
Sample Volume:	6.2 µL
Reagent 1 Volume:	34.7 µL
Reagent 2 Volume:	34.7 µL
Reaction Time:	3.8 minutes
Test Temperature:	37° C
Wavelength:	540 & 700 nm
Type of measurement:	Bichromatic endpoint

NOTE: In the event that the test system becomes inoperable, notify supervision or designee for further direction. Patient specimens must be stored in a manner that maintains the integrity of the specimen.

9. CALCULATIONS

The instrument automatically calculates the concentration of Total Protein in g/dL.

Albumin/globulin (A/G) ratio is given whenever the Total Protein and Albumin are ordered at the same time. Since the total protein value is elevated by the inclusion of fibrinogen in plasma specimens, the *Total Protein is corrected for this calculation*. Therefore, the formula is as follows:

(Total Protein in g/dL – 0.3g/dL) – Albumin (g/dL) = the Globulin Proteins g/dL

$$\frac{\text{Albumin (g/dL)}}{\text{Globulin Proteins g/dL}} = \text{A / G ratio}$$

10. REPORTING RESULTS AND REPEAT CRITERIA

10.1 Interpretation of Data

None required

10.2 Rounding

No rounding is necessary. Instrument reports results up to one decimal point.

10.3 Units of Measure

g/dL

10.4 Clinically Reportable Range (CRR)

0.0 – 36.0 g/dL

10.5 Review Patient Data

Each result is reviewed for error messages. Refer to the Dimension Vista system manual "Error messages" section for troubleshooting. Resolve any problems noted before issuing patient reports.

10.6 Repeat Criteria and Resulting

All repeats must replicate the original result within the total allowable error (TEa) of the assay. Refer to TEa listing for specific information.

Values that fall within the AMR or CRR may be reported without repeat. Values that fall outside these ranges must be repeated.

IF the result is …	THEN…
0.0 g/dL	Assure there is sufficient sample devoid of bubbles, cellular debris, and/or fibrin clots. Report as: 0.0 g/dL
≥ 12.0 g/dL	**On Board Automated Dilution:** Results ≥ 12.0 g/dL will automatically have repeat testing performed into the instrument using dilution factor of 2. No multiplication is necessary.
> 24.0 g/dL	**Manual Dilution:** Using the primary tube, make the smallest dilution possible to bring the raw data within the AMR. Maximum allowable dilution: x 3 **DILUENT**: Water Enter dilution factor as a whole number. Re-assay. Readout is corrected for dilution.
> 36.0 g/dL	If the recommended dilution does not give results within the clinically reportable range, report as: "> 36.0 g/dL-REP" Bring to the attention of your supervisor prior to releasing result.

Message	Code
Verified by repeat analysis	Append –REP to the result.

11. EXPECTED VALUES

11.1 Reference Ranges

Serum / Plasma:

Age	Female	Male
Adult (>19 years):	6.4 – 8.2 g/dL	6.4 – 8.2 g/dL
Pediatric:		
10 – 19 years	6.4 - 8.6	6.4 - 8.6
7 – 9 years	6.3 - 8.1	6.3 - 8.1
1 – 6 years	6.0 - 7.8	6.0 - 8.0
6 – 11 months	4.6 - 7.8	4.2 - 7.9
61 – 180 days	4.0 - 7.6	4.0 - 7.0
0 – 60 days	3.6- 7.0	4.0 - 7.6

Body Fluid:
Exudates > 3.0 g/dL
Transudates < 3.0 g/dL

A/G ratio: 1.1 - 2.0

11.2 Critical Values

None established

11.3 Standard Required Messages

None established

12. CLINICAL SIGNIFICANCE

Measurements of total protein are used in the diagnosis and treatment of a variety of diseases involving the liver, kidney or bone marrow as well as metabolic or nutritional disorders.

13. PROCEDURE NOTES

- **FDA Status:** FDA Approved/cleared
- **Validated Test Modifications:** None

The instrument reporting system contains error messages to warn the operator of specific malfunctions. Any report slip containing such error messages should be held for follow-up. Refer to your Dimension Vista Operator's Guide.

The expected maximum observed standard deviations for repeatability using n = 5 replicates at the following total protein concentrations are:

TP Concentration	Acceptable S.D. Maximum
3.7 g/dL	0.4 g/dL
8.3 g/dL	0.8 g/dL

14. LIMITATIONS OF METHOD

14.1 Analytical Measurement Range (AMR)

0.0 – 12.0 g/dL

14.2 Precision

Material	Mean g/dL	Standard Deviation (%CV)	
		Repeatability	Within-Lab
Multiqual Control			
Level 1	3.7	0.1 (2.9)	0.1 (3.2)
Level 2	8.3	0.2 (2.3)	0.2 (2.5)

14.3 Interfering Substances

Dextran 40 increased TP results by 38% at 7 g/dL of total protein.

HIL Interference:
The TP method was evaluated for interference according to CLSI/NCCLS EP7-A2. Bias, defined as the difference between the control sample (does not contain interferent) and the test sample (contains interferent), is shown in the table below. Bias exceeding 10% is considered “interference”.

Substance tested	Substance Concentration	TP g/dL	Bias %
Hemoglobin (hemolysate)	1000 mg/dL	6.7	<10
Bilirubin (unconjugated)	20 mg/dL	6.9	<10
Bilirubin (conjugated)	20 mg/dL	7	<10
Lipemia Intralipid®	1000 mg/dL	6.2	<10

14.4 Clinical Sensitivity/Specificity/Predictive Values

Not available

15. SAFETY

Refer to your local and corporate safety manuals and Safety Data Sheet (SDS) for detailed information on safety practices and procedures and a complete description of hazards.

TP Flex® Reagent Cartridge may corrosive to metals. Very toxic to aquatic life. Harmful to aquatic life with long lasting effects. Avoid release to the environment.

16. RELATED DOCUMENTS

1. Dimension Vista® Clinical Chemistry System Operator's Manual
2. Dimension Vista® Calibration/Verification Procedure
3. Dimension Vista® Cal Accept Guidelines
4. Dimension Vista® Calibration summary
5. Dimension Vista® Sample Processing, Startup and Maintenance procedure
6. Laboratory Quality Control Program
7. QC Schedule for Siemens Dimension Vista®
8. Laboratory Safety Manual
9. Safety Data Sheets (SDS)
10. Dimension Vista® Limits Chart (AG.F200)
11. Quest Diagnostics Records Management Procedure
12. Dimension Vista® System Error Messages Chart
13. Centrifuge Use, Maintenance and Functions Checks (Lab policy)
14. Hemolysis, Icteria and Lipemia Interference (Lab policy)
15. Repeat Testing Requirement (Lab policy)
16. Current Allowable Total Error Specifications
    at http://questnet1.qdx.com/Business_Groups/Medical/qc/docs/qc_bpt_tea.xls
17. Current package insert TP Flex® Reagent Cartridge K1073

17. REFERENCES

1. Ghoshal, Amit K. and Soldin, Steven J., Evaluation of the Dade Behring Dimension® RxL: Integrated chemistry system-pediatric reference ranges. Clinica Chimica Acta 2003; 331:144.
2. Package Insert, TP Flex® Reagent Cartridge K1073, Siemens Healthcare Diagnostics Inc., 05/11/2015.
3. Package Insert, CHEM 4 CAL, Siemens Healthcare Diagnostics Inc., 03/2008.
4. Package Insert, Liquid Assayed Multiqual® Chemistry Controls, Bio-Rad Laboratories, 09/2015.
5. Quest Diagnostics SOP ID 300SA373, Total Protein, Serum and Fluid.

18. REVISION HISTORY

Version	Date	Section	Reason	Reviser	Approval
000	3/27/14		Update owner	L Barrett	R SanLuis
000	3/27/14	5.2	Remove 31 day stability	A Chini	R SanLuis
000	3/27/14	16	Update titles	L Barrett	R SanLuis
000	3/27/14	Footer	Version # leading zero's dropped due to new EDCS in use as of 10/7/13.	L Barrett	R SanLuis
1	2/13/17	Header	Add WAH	L Barrett	R SanLuis
1	2/13/17	3.2	Specify anticoagulant, remove specimen onboard stability	L Barrett	R SanLuis
1	2/13/17	4,5,6	Remove individual section labeling instructions and add general one	L Barrett	R SanLuis
1	2/13/17	6.1, 6.2	Update QC material and storage	L Barrett	R SanLuis
1	2/13/17	6.4, 6.6	Replace LIS with Unity Real Time	L Barrett	R SanLuis
1	2/13/17	7.2	Change freezer upper limit to -50C	L Barrett	R SanLuis
1	2/13/17	10.5	Move patient review from section 6	L Barrett	R SanLuis
1	2/13/17	11.1	Add A/G ratio range	L Barrett	R SanLuis
1	2/13/17	15	Update to new standard wording, add warning	L Barrett	R SanLuis
1	2/13/17	17	Update QC product and PI dates	L Barrett	R SanLuis

19. ADDENDA

None