

TRAINING UPDATE

Lab Location: SGMC & WOMC
Department: Core Lab

Date Distributed: 9/19/2020
Due Date: 9/30/2020
Implementation: 9/29/2020

DESCRIPTION OF PROCEDURE REVISION

Name of procedure:								
Urine Buprenorphine Screen by Dimension Vista® System SGMC.C2004 v2 Amikacin by Siemens Immunoassay on Dimension Vista® System SGMC.C2001 v2 Vista Reagent QC Handoff Log AG.F538.1								
Description of change(s):								
SOPs								
<table border="1"><thead><tr><th>Section</th><th>Reason</th></tr></thead><tbody><tr><td>4.2</td><td>Added tech code to flex labeling requirement</td></tr><tr><td>6.3</td><td>Added QC required after every new flex</td></tr><tr><td>16</td><td>Added QC handoff log</td></tr></tbody></table>	Section	Reason	4.2	Added tech code to flex labeling requirement	6.3	Added QC required after every new flex	16	Added QC handoff log
Section	Reason							
4.2	Added tech code to flex labeling requirement							
6.3	Added QC required after every new flex							
16	Added QC handoff log							
LOG – this is new, similar to Coag log used to record reagent prep and placement								
The revised SOPs and FORM will be implemented Sept 29, 2020								

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

Technical SOP

Title	Urine Buprenorphine Screen by Dimension Vista® System	
Prepared by	Ashkan Chini	Date: 5/18/2020
Owner	Robert SanLuis	Date: 5/18/2020

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

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1. TEST INFORMATION

Assay	Method/Instrument	Test Code
Urine Buprenorphine, Qualitative	Dimension Vista® System	UBUP

Synonyms/Abbreviations
Subutex®, BUP Included in Urine Drug Screen (UDRGT) and Urine Drug Screen with TCA(UDRGW)

Department
Chemistry

2. ANALYTICAL PRINCIPLE

The Emit II Plus Buprenorphine Assay is a homogeneous enzyme immunoassay technique used for the analysis of specific compounds in human urine. The assay is based on competition between drug in the specimen and drug labeled with the recombinant glucose-6-phosphate dehydrogenase (rG6PDH) for antibody binding sites. Enzyme activity decreases upon binding to the antibody, so the drug concentration in the specimen can be measured in terms of enzyme activity. Active enzyme converts nicotinamide adenine dinucleotide (NAD) to NADH in the presence of glucose-6-phosphate (G6P), resulting in an absorbance change that is measured spectrophotometrically. Endogenous serum G6PDH does not interfere because the coenzyme NAD functions only with the bacterial (*Leuconostoc mesenteroides*) enzyme employed in the assay.

3. SPECIMEN REQUIREMENTS

3.1 Patient Preparation

Component	Special Notations
Fasting/Special Diets	N/A
Specimen Collection and/or Timing	Freshly voided urine specimens should be used for testing.
Special Collection Procedures	No additives or preservatives are needed. Adulteration of the urine specimen may cause erroneous results. If adulteration is suspected, obtain a fresh specimen. Urine specimens should be handled and treated as if they are potentially infected. Preferred method is the Urine Collection Kit with specimen transferred to Urine Chemistry Collection Tube (yellow top).

Component	Special Notations
Other	If Urine Collection Kit is not used, submit to Laboratory within 2 hours of collection.

3.2 Specimen Type & Handling

Criteria	
Type -Preferred -Other Acceptable	Urine None
Collection Container	Urine Collection Kit or sterile container
Volume - Optimum - Minimum	15 mL 2 mL
Transport Container and Temperature	Urine Chemistry Collection Tube (yellow top) or container at room temperature.
Stability & Storage Requirements	Room Temperature: 5 days
	Refrigerated: 5 days
	Frozen: If storage longer than 5 days is required.
Timing Considerations	Deliver specimens to laboratory immediately.
Unacceptable Specimens & Actions to Take	Specimens that are unlabeled, improperly labeled, or those that do not meet the stated criteria are unacceptable. Samples in Urine Analysis Preservative Tubes are NOT acceptable. 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	Turbidity: Centrifuge turbid samples before analysis. Frozen samples must be thawed and mixed thoroughly prior to analysis. Avoid repeat freeze/thaw cycles.
Other Considerations	Boric acid should not be used as a preservative. Plastic transfer pipettes should NOT be used for delivering patient specimen to sample cup.

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.

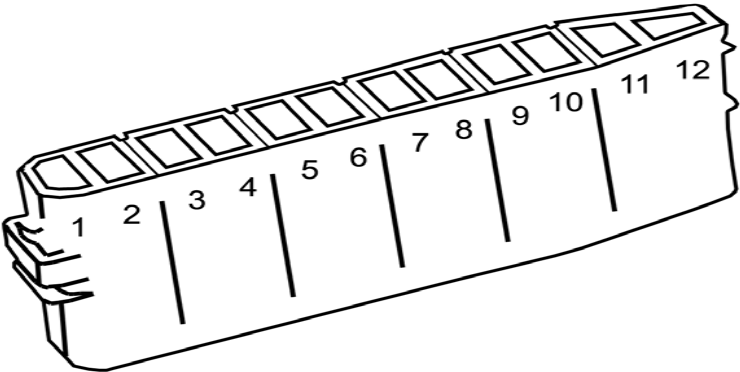
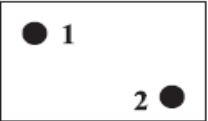
4. REAGENTS

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
Buprenorphine Assay (consists of Antibody/Substrate Reagent 1 and Enzyme Reagent 2)	Siemens, Syva® Emit II Plus, Cat. No. 10720048
Empty Flex Reagent Cartridge	Siemens, Cat. No. 10445218

4.2 Reagent Preparation and Storage

Reagents	Antibody/Substrate Reagent 1 & Enzyme Reagent 2
Container	Plastic vial, Flex Reagent Cartridge
Storage	Stored at 2–8°C, upright and with screw caps tightly closed
Stability	<ul style="list-style-type: none"> • Antibody/Substrate Reagent 1 & Enzyme Reagent 2: Stable until the expiration date printed on the label • Pre-filled Flex Reagent Cartridge: When taped and stored at 2–8°C, it will remain stable for 7 days. Remove the tape before loading the Flex reagent cartridge on the analyzer. • Pre-filled Flex Reagent Cartridge: When loaded on the Dimension Vista, it will remain stable for 24 hours.
Preparation	<p>Both Antibody/Substrate Reagent 1 and Enzyme Reagent 2 are ready for use. No preparation is required.</p> <p>The wells of an empty Flex Reagent Cartridge are identified as shown below.</p>  <p>The Flex well must be vented before filling with reagent. To vent a Flex well, puncture the film at a corner of the well (see picture below). Care must be taken to minimize the size of the vent; do not tear the film.</p>  <p>Transfer reagent into an empty, vented Flex Reagent Cartridge</p>

	<p>according to the table below. Try to avoid bubbles as much as possible. Label the Flex Reagent Cartridge with reagent name, lot number, date prepared, expiration date, and tech code.</p> <table border="1" style="margin-left: auto; margin-right: auto;"> <thead> <tr> <th>Reagent</th> <th>Wells</th> <th>Volume per Well</th> <th>Tests per Well</th> </tr> </thead> <tbody> <tr> <td>Reagent 2</td> <td>1 and 2</td> <td>0.8 mL</td> <td>10</td> </tr> <tr> <td>Reagent 1</td> <td>11 and 12</td> <td>1.2 mL</td> <td>10</td> </tr> </tbody> </table>	Reagent	Wells	Volume per Well	Tests per Well	Reagent 2	1 and 2	0.8 mL	10	Reagent 1	11 and 12	1.2 mL	10
Reagent	Wells	Volume per Well	Tests per Well										
Reagent 2	1 and 2	0.8 mL	10										
Reagent 1	11 and 12	1.2 mL	10										

4.3 Loading Pre-filled Flex Reagent Cartridge on Dimension Vista

Note: Load one Empty flex on board at a time. Since there is no way to differentiate between empty flexes on board, operator must load one empty flex and identify it before loading the next empty flex.

Load the flex on board. From the Home Page go to **Set Up – Inventory – Reagents** – Select “**Empty**” – Identify the method and lot number – **Finish**

5. CALIBRATORS/STANDARDS

5.1 Calibrators/Standards Used

Calibrator	Supplier and Catalog Number
Emit® Calibrator/Control Level 0	Siemens Syva® Emit, Cat. No. 10445406
Emit® II Plus Specialty Drug Calibrator/Control Levels 1, 2, 3, & 4	Siemens Syva® Emit, Cat. No. 10720049 - 10720052

5.2 Calibrator Preparation and Storage

Calibrator	Emit® Calibrator/Control Level 0 and Emit® II Plus Specialty Drug Calibrator/Control Levels 1, 2, 3, & 4
Preparation	Calibrator is ready for use. No preparation is required.
Storage/Stability	<ul style="list-style-type: none"> • Store at 2-8°C, upright • If stored as directed, calibrators are stable until the expiration date printed on the label.

5.3 Calibration Parameter

Criteria	Special Notations
Reference Material	Emit® Calibrator/Control Emit® II Plus Specialty Drug Calibrator/Control
Assay Range	0 – 25 ng/mL

Suggested Calibration Level	<ul style="list-style-type: none"> • Level 0 = 0 ng/mL • Level 1 = 2.5 ng/mL • Level 2 = 5 ng/mL • Level 3 = 15 ng/mL • Level 4 = 25 ng/mL
Frequency	<ul style="list-style-type: none"> • Every new reagent cartridge lot. • Every 14 days for any one lot. • When major maintenance is performed on the analyzer. • When control data indicates a significant shift in assay.
Calibration Scheme	5 Levels

5.4 Calibration Procedure

Manual Calibration:

1. Verify that reagent is in inventory on the instrument.
2. From the Home Page select **Advanced – Calibration – Calibrations by Lot** – Select “**XBUP**” – **Order Calibration** – Designate a specimen rack and scan it - **OK**
3. Pour calibrators in Sample Cups and load them as described below:
 - Calibrator 0 on rack position 1
 - Calibrator 1 on rack position 2
 - Calibrator 2 on rack position 3
 - Calibrator 3 on rack position 4
 - Calibrator 4 on rack position 5
4. After loading the rack on the instrument, 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
Specialty Drug Controls Negative	Siemens Syva® Emit II Plus, Cat. No. 10718700
Specialty Drug Controls Positive	Siemens Syva® Emit II Plus, Cat. No. 10718701

6.2 Control Preparation and Storage

Control	Syva Emit II Plus Controls Negative and Positive
Preparation	The controls are ready for use. No preparation is required. <ul style="list-style-type: none"> • Aliquot 2 mL into screw top vials • Print labels from the Vista and label each vial • Immediately load onto the instrument for 2–8°C storage
Storage/Stability	<ul style="list-style-type: none"> • Store at 2–8°C • Stable opened or unopened in the original container until the expiration date printed on the vial. • On board Dimension Vista, QC vials will remain stable for 7 days.

6.3 Frequency

Analyze all levels of QC material **immediately after loading a new flex cartridge***, after every calibration and each day of testing (notated on the QC frequency sheets posted on the instruments).

*** Document the new flex reagent and QC assessment on the Vista Reagent QC Handoff Log.**

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 <ul style="list-style-type: none"> • 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: <ul style="list-style-type: none"> • 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 <u>reanalyzed according to the Laboratory QC Program</u>. 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.

Step	Action
	<ul style="list-style-type: none"> • Corrective action documentation must follow the Laboratory Quality Control Program.
4	<p>Review of QC</p> <ul style="list-style-type: none"> • 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
- Screw top vials, 3mL

8. PROCEDURE

XBUP Flex® reagent cartridge Cat. No. 10720048 is required to perform this test.

Buprenorphine 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 10.6 “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.

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

None

10. REPORTING RESULTS AND REPEAT CRITERIA

10.1 Interpretation of Data

None required

10.2 Rounding

N/A

10.3 Units of Measure

N/A

10.4 Clinically Reportable Range (CRR)

N/A

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

Specimens that give an “Abnormal Reaction” message must be repeated.

If the result is Positive **and** the patient’s location is L&D (Labor and Delivery), DI will flag the result to be held for repeat. Repeat the test, if possible run repeat on another analyzer:

- If repeat is positive, report as POSITIVE
- If repeat is negative, run QC to verify instrument performance. If QC is within range on the same reagent lot as used for patient run, rerun patient sample a third time. Report result that duplicated (2 of 3 results).

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

11. EXPECTED VALUES

11.1 Reference Ranges

None detected

11.2 Critical Values

None established

11.3 Standard Required Messages

The following comment is automatically added to the report by the LIS when a urine Buprenorphine test is ordered:

This is a screening assay. Buprenorphine is detected in concentration at or above 5 ng/mL

The following comment is automatically added to the report by the LIS when a urine drug screen is ordered:

The drug of abuse panel is a screening assay. It detects the following drugs of abuse in concentrations at or above the concentrations listed below.

Phencyclidine	25 ng/mL
Benzodiazepines	200 ng/mL
Cocaine	300 ng/mL
Amphetamines	1000 ng/mL
THC	50 ng/mL
Opiates	300 ng/mL
Barbiturates	200 ng/mL
Methadone	300 ng/mL
Buprenorphine	5 ng/mL

The ingestion of certain herbal or plant products containing Ephedra or its metabolites may cause false positive amphetamine/metamphetamine results.

This test is for medical screening purposes ONLY. For confirmation a separate order for Gas Chromatography by Mass Spectrophotometry (GCMS) is required.

12. CLINICAL SIGNIFICANCE

Buprenorphine is a semi-synthetic opioid analgesic derived from thebaine, a component of the opium poppy *Papaver somniferum*. Buprenorphine structurally resembles morphine, but has both antagonist and agonist properties. Buprenorphine is a schedule III drug. The Food and Drug Administration (FDA) has approved the use of Suboxone, which contains buprenorphine for treatment of opiate dependency in the US. Under the US Drug Abuse Treatment Act of 2000 (DATA) buprenorphine can be prescribed in a physician's office for treatment of opiate dependency.

It has been shown that buprenorphine has abuse potential and may itself cause dependency. It produces typical opioid effects and side effects such as euphoria and respiratory depression.

Buprenorphine is metabolized in the human liver primarily by N-dealkylation to pharmacologically active norbuprenorphine, which along with the parent compound is conjugated to form buprenorphine glucuronide and norbuprenorphine glucuronide.

13. PROCEDURE NOTES

- **FDA Status:** FDA Approved/Modified
- **Validated Test Modifications:** Removed pH testing per communication from manufacturer

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.

14. LIMITATIONS OF METHOD

14.1 Analytical Measurement Range (AMR)

Qualitative Assay: 0.7 – 25 ng/mL (for 5 ng/mL cutoff)

14.2 Precision

Urine Pool (ng/mL)	% of Cutoff	# of Results	Repeatability Results	Within-Laboratory Results
1.25	-75%	80	80 Negative	80 Negative
8.75	+75%	80	80 Positive	80 Positive

14.3 Interfering Substances

None

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 Function Checks (Lab policy)
14. Specimen Acceptability Requirements (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 XBUP Flex® Reagent Cartridge 10720048
18. Vista Reagent QC Handoff Log (AG.F538)

17. REFERENCES

1. Package Insert, XBUP Flex® Reagent Cartridge 10720048, Siemens Healthcare Diagnostics Inc., 05/2019.
2. Package Insert, Emit Calibrator, Siemens Healthcare Diagnostics Inc., 05/2019.
3. Package Insert, Emit II Plus Specialty Drug Calibrator, Siemens Healthcare Diagnostics Inc., 05/2019.
4. Package Insert, Emit II Plus Specialty Drug Control, Bio-Rad Laboratories, 05/2019.

18. REVISION HISTORY

Version	Date	Section	Reason	Reviser	Approval
1	9/2/20	4.2	Added tech code to flex labeling	L Barrett	R SanLuis
1	9/2/20	6.3	Added QC required after every new flex	L Barrett	R SanLuis
1	9/2/20	16	Added QC handoff log	L Barrett	R SanLuis

19. ADDENDA

Validated Test Modification statement from manufacturer

From: Brodbeck, Beate (H USA)
Sent: Thursday, June 24, 2010 3:54 PM
To: SanLuis, Robert; Mcmillan, Wendell R
Subject: pH for urine DAU samples

Hi Robert and Wendell,

Below is the response regarding your inquiry of pH testing for drugs of abuse urines.

Beate Brodbeck

Chemistry Instrument Specialist - Western Maryland

Siemens Healthcare Diagnostics
<blocked::http://www.siemens.com/diagnostics>

C: 410-370-4382 | VM: 800-948-3234 x-2684

beate.brodbeck@siemens.com

From Kevin Mulrooney:

pH correction of urine samples prior to running the Drugs of Abuse assays on Dimension is not an absolute requirement. The Dimension IFU's all say that the acceptable pH range is 5-8. The Syva Emit IFU's for the same tests all say that the acceptable pH range is 3-11, except for THC (pH range 4.5-8). The assays all work at pH 3-11. The vast majority of urines will fall in this range. THC is an exception in that at acid pH <4.5, THC recovery is decreased, and at basic pH >8, THC recovery is increased. When the Dimension IFU's were written, I suppose the decision was made to standardize the pH acceptable range to the most narrow (THC). Dimension customers can run the DAT's without checking pH, but there is a slight chance of inaccuracy with THC. Urine pH outside the 5-8 range is not common, either.

Hope this helps. We don't have this in a formal document, but you can share this information with your customer.

Regards,

Customer Care.
With you every step of the way.

Kevin Mulrooney
Kevin Mulrooney
Staff Product Support Specialist
Global Product Support
Siemens Healthcare Diagnostics
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Technical SOP

Title	Amikacin by Siemens Immunoassay on Dimension Vista® System	
Prepared by	Ashkan Chini	Date: 4/13/2020
Owner	Robert SanLuis	Date: 4/13/2020

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

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1. TEST INFORMATION

Assay	Method/Instrument	Test Code
Amikacin, Peak	Siemens Immunoassay on Dimension Vista® System	AMIKP
Amikacin, Trough		AMIKT
Amikacin, Random		AMKR

Synonyms/Abbreviations
Amikin

Department
Chemistry

2. ANALYTICAL PRINCIPLE

The Syva Emit Amikacin assay is a homogeneous enzyme immunoassay technique used for the analysis of specific compounds in biological fluids. The assay is based on competition between drug in the sample and drug labeled with the enzyme glucose-6-phosphate dehydrogenase (G6PDH) for antibody binding sites. Enzyme activity decreases upon binding to the antibody, so the drug concentration in the sample can be measured in terms of enzyme activity. Active enzyme converts oxidized nicotinamide adenine dinucleotide (NAD) to NADH, resulting in an absorbance change that is measured spectrophotometrically. Endogenous serum G6PDH does not interfere, because the coenzyme functions only with the bacterial (*Leuconostoc mesenteroides*) enzyme employed in the assay.

3. SPECIMEN REQUIREMENTS

3.1 Patient Preparation

Component	Special Notations
Fasting/Special Diets	N/A
Specimen Collection and/or Timing	Trough: Collect immediately before dose (within 30 minutes) Peak: Collect at end of a 60 minute IV infusion, or 30 minutes after end of 30 minute infusion, or 60 minutes after IM dose
Special Collection Procedures	N/A
Other	N/A

3.2 Specimen Type & Handling

Criteria	
Type	Serum
-Preferred	
-Other Acceptable	None

Criteria	
Collection Container	Serum: Red top tube, Serum separator tube (SST)
Volume - Optimum	1.0 mL
- Minimum	0.5 mL
Transport Container and Temperature	Collection container or Plastic vial at room temperature
Stability & Storage Requirements	Room Temperature: 7 days
	Refrigerated: 7 days
	Frozen: 7 days
Timing Considerations	Serum 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.

Note: Reagents A and B and calibrators are provided as a matched set. They should not be interchanged with components of kits with different lot numbers.

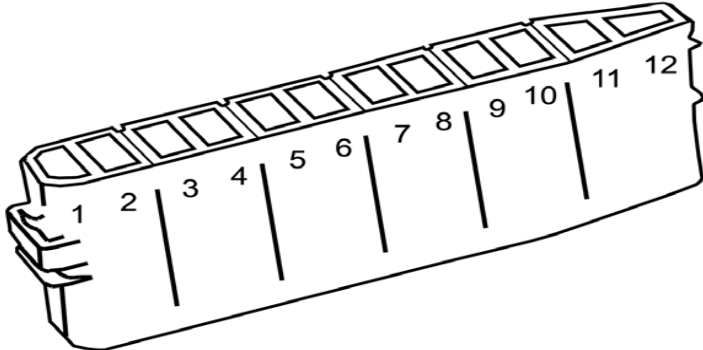
4.1 Reagent Summary

Reagents	Supplier & Catalog Number
Amikacin	Siemens, Syva® Emit Amikacin Assay, Cat. No. 10445383
Empty Flex Reagent Cartridge	Siemens, Cat. No. 10445218


4.2 Reagent Preparation and Storage

Reagent	<ul style="list-style-type: none"> • Enzyme Reagent A • Antibody/Substrate Reagent B • Buffer Concentrate
Container	Glass vial, Flex Reagent Cartridge
Storage	Stored at 2–8°C
Stability	<ul style="list-style-type: none"> • Unopened: stable until expiration date printed on each vial • Working Reagent A remains stable at 2-8°C for 12 weeks • Working Reagent B remains stable at 2-8°C for 12 weeks • Buffer Solution remains stable at 20-25°C for 12 weeks • Pre-filled Flex Reagent Cartridge: When loaded on the Dimension Vista, it will remain stable for 48 hours.
Reagent A & B Reconstitution	<p>Reconstitute both Enzyme Reagent A and Antibody/Substrate Reagent B using 6 mL of reagent grade water.</p> <ul style="list-style-type: none"> • Gently swirl the vial to dissolve the contents. • Allow to equilibrate at 20-25°C for the minimum of 2 hours.
Working Buffer Solution Preparation	<p>Make a 1:15 dilution of Buffer Concentrate using reagent grade water as the diluent.</p> <ul style="list-style-type: none"> • Mix 4 mL of Buffer Concentrate with 56 mL of reagent grade water; invert gently to ensure it is homogenous. • Prepare two different Buffer Solutions, one for Working Reagent A and one for Working Reagent B.
1:9 Working A & B Reagent Preparation	<p>Note: Use sterile containers labeled with reagent name, lot number, and expiration date for the following steps:</p> <ol style="list-style-type: none"> 1. Working Reagent A: Make a 1:9 dilution of reconstituted Enzyme Reagent A using Buffer Solution as the diluent. Invert both the reconstituted Enzyme Reagent A vial and Buffer Solution gently to ensure they are homogenous. Mix 6 mL of reconstituted Enzyme Reagent A with 48 mL of Buffer Solution; invert gently to ensure it is homogenous. 2. Working Reagent B: Make a 1:9 dilution of reconstituted Antibody/Substrate Reagent B using Buffer Solution as the diluent. Invert both the reconstituted Antibody/Substrate Reagent B vial and Buffer Solution gently to ensure they are homogenous. Mix 6 mL of reconstituted Antibody/Substrate Reagent B with 48 mL of Buffer Solution; invert gently to ensure it is homogenous. <p>Note: After preparation, allow the working reagents A and B to equilibrate at 2–8°C for one hour before proceeding to the next step.</p>

The wells of an empty Flex Reagent Cartridge are identified as shown below.



The Flex well must be vented before filling with reagent. To vent a Flex well, puncture the film at a corner of the well (see picture below). Care must be taken to minimize the size of the vent; do not tear the film.



Transfer reagent into an empty, vented Flex Reagent Cartridge according to the table below. Try to avoid bubbles as much as possible. Label the Flex Reagent Cartridge with reagent name, lot number, date prepared, expiration date, and tech code.

Reagent	Wells	Volume per Well	Tests Per Well
Reagent B	1 and 2	1.2 mL	10
Reagent A	11 and 12	1.2 mL	10

4.3 Loading Pre-filled Flex Reagent Cartridge on Dimension Vista

Note: Load one Empty flex on board at a time. Since there is no way to differentiate between empty flexes on board, operator must load one empty flex and identify it before loading the next empty flex.

Load the flex on board. From the Home Page go to **Set Up – Inventory – Reagents** – Select “**Empty**” – Identify the method and lot number - **Finish**

5. CALIBRATORS/STANDARDS

5.1 Calibrators/Standards Used

Calibrator	Supplier and Catalog Number
Amikacin Calibrator	Siemens, Syva® Emit Amikacin Assay, Cat. No. 10445383

Note: Reagents A and B and calibrators are provided as a matched set. They should not be interchanged with components of kits with different lot numbers.

5.2 Calibrator Preparation and Storage

Calibrator	Amikacin Calibrator
Preparation	Reconstitute each calibrator vial using 1 mL of reagent grade water; gently swirl the vial to dissolve the contents. Allow to equilibrate at 20-25°C for the minimum of 2 hours. Note: Calibrator Level 0 is not required for calibration. It is used as a diluent for samples with values > 50 µg/mL. Refer to section 10.6.
Storage/Stability	<ul style="list-style-type: none"> • Unopened vials: stable until the expiration date on the vial • Reconstituted: stable at 2-8°C for 12 weeks

5.3 Calibration Parameter

Criteria	Special Notations
Reference Material	Emit® Amikacin Calibrator
Assay Range	2.5 – 50 µg/mL
Suggested Calibration Level	<ul style="list-style-type: none"> • Level 1 = 2.5 µg/mL • Level 2 = 5 µg/mL • Level 3 = 10 µg/mL • Level 4 = 20 µg/mL • Level 5 = 50 µg/mL
Frequency	<ul style="list-style-type: none"> • Every new reagent cartridge lot. • Every 14 days for any one lot • When major maintenance is performed on the analyzer. • When control data indicates a significant shift in assay.
Calibration Scheme	5 Levels

5.4 Calibration Procedure

Note: Calibrator Level 0 is not needed for calibration. However, it is used as a diluent for samples with values > 50 µg/mL. Refer to section 10.6.

1. Verify that reagent is in inventory on the instrument.

2. From the Home Page select **Advanced – Calibration – Calibrations by Lot** – Select “**XAMIK**” – **Order Calibration** – Designate a specimen rack and scan it - **OK**
3. Pour calibrators in Sample Cups and load them as described below:
 - Level 1 (2.5 µg/mL) on rack position 1
 - Level 2 (5 µg/mL) on rack position 2
 - Level 3 (10 µg/mL) on rack position 3
 - Level 4 (20 µg/mL) on rack position 4
 - Level 5 (50 µg/mL) on rack position 5
4. After loading the rack on the instrument, 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
Liquichek™ Immunoassay Plus Control Levels 1, 2 and 3	Bio-Rad Laboratories Cat. No. 267, 268 and 269

6.2 Control Preparation and Storage

Control	Liquichek Immunoassay Plus Controls, Level 1, 2 and 3
Preparation	Allow the vials to stand at room temperature (18 – 25°C) until completely thawed. Gently swirl the vials several times to ensure homogeneity and immediately load onto instrument.
Storage/Stability	Thawed: all analytes will be stable for 4 days at 2 – 8°C. Frozen: stable until the expiration date at -20 to -50°C.

6.3 Frequency

Analyze all levels of QC material **immediately after loading a new flex cartridge***, after every calibration and each day of testing (notated on the QC frequency sheets posted on the instruments).

*** Document the new flex reagent and QC assessment on the Vista Reagent QC Handoff Log.**

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	<p>Run Rejection Criteria</p> <ul style="list-style-type: none"> 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	<p>Corrective Action:</p> <ul style="list-style-type: none"> 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 <u>reanalyzed according to the Laboratory QC Program</u>. 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	<p>Review of QC</p> <ul style="list-style-type: none"> 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
- Reagent grade water

8. PROCEDURE

XAMIK Flex® reagent cartridge Cat. No. 10445383 is required to perform this test.

Amikacin 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.
4.	Follow protocol in Section 10.6 “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.

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 Amikacin in µg/mL.

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

µg/mL

10.4 Clinically Reportable Range (CRR)

2.5 – 100.0 µg/mL

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 below or within the AMR or CRR may be reported without repeat. Values that exceed the upper ranges must be repeated.

IF the result is ...	THEN...
< 2.5 µg/mL	Assure there is sufficient sample devoid of bubbles, cellular debris, and/or fibrin clots. Report as: < 2.5 µg/mL
≥ 50.0 µg/mL	Manual Dilution: Using the primary tube, make the smallest dilution possible to bring the raw data within the AMR. Maximum allowable dilution: x 2 Diluent: Emit® Amikacin Calibrator 0 Enter dilution factor as a whole number.
> 100.0 µg/mL	If the recommended dilution does not give results within the clinically reportable range, report as: “> 100.0 µg/mL -REP” Bring to the attention of Tech in Charge (TIC) or Group Lead to check for integrity issues prior to release of results.

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

11. EXPECTED VALUES

11.1 Reference Ranges

Random: None established
 Peak: 20.0 – 30.0 µg/mL
 Trough: 4.0 – 8.0 µg/mL

11.2 Critical Values

Random: > 30.0 µg/mL
Peak: > 30.0 µg/mL
Trough: > 8.0 µg/mL

11.3 Standard Required Messages

None established

12. CLINICAL SIGNIFICANCE

Monitoring Amikacin concentrations in serum is the most effective means of ensuring adequate therapy. Amikacin concentration in serum correlates better with antibacterial activity than does dosage. A standard dose of amikacin does not always yield a predictable serum level because the drug's concentration also depends on the patient's volume of distribution and on drug elimination. These factors are influenced by the mode of administration, the volume of extracellular fluid, renal function, and physiological changes during therapy. Patients with impaired renal function, dialysis patients, burn patients, and neonatal or elderly patients should be monitored closely. Exposure to high concentrations for a prolonged period may cause renal impairment or ototoxicity.

13. PROCEDURE NOTES

- **FDA Status:** FDA Approved/Modified
- **Validated Test Modifications:** Specimen stabilities have been modified from the package insert based on in-house stability studies performed at Quest Diagnostics. (per *Amikacin by Siemens Immunoassay on the Beckman Coulter/Olympus AU 400/600/5400/2700 Series*, SOP ID QDTX722)

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.

14. LIMITATIONS OF METHOD

14.1 Analytical Measurement Range (AMR)

2.5 – 50.0 µg/mL

14.2 Precision

Within Run	Number of Replicates	Mean µg/mL	Coefficient of Variation (%)
1	20	9.2	8.6
2	20	9.9	3

14.3 Interfering Substances

Patient samples containing kanamycin will cause significant elevation of amikacin results.

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.

The reagent contains Sodium azide; 2-methyl-3(2h)-isothiazolone, hydrochloride. Harmful if swallowed or in contact with skin. Causes serious eye irritation. Causes skin irritation. May cause allergy or asthma symptoms or breathing difficulties if inhaled. May cause an allergic skin reaction.

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 Function Checks (Lab policy)
14. Specimen Acceptability Requirements (Lab policy)
15. Repeat Testing Requirement (Lab policy)
16. Therapeutic Drug Monitoring (Chemistry SOP)
17. Current Allowable Total Error Specifications at
http://questnet1.qdx.com/Business_Groups/Medical/qc/docs/qc_bpt_tea.xls
18. Current package insert Siemens Syva® Emit Amikacin Assay 10445383
19. Vista Reagent QC Handoff Log (AG.F538)

17. REFERENCES

1. Quest Diagnostics SOP, Amikacin by Siemens Immunoassay on the Beckman Coulter/Olympus AU 400/600/5400/2700 Series, SOP ID QDTX722.
2. Package Insert, Syva® Emit Amikacin Assay 10445383, Siemens Healthcare Diagnostics Inc., 05/2015.

3. Emit® Amikacin Application Sheet Dimension Vista® Systems with software version 3.2.1 or higher, Siemens Healthcare Diagnostics Inc., 03/28/2018.
4. Package Insert, Liquichek Immunoassay Plus, Bio-Rad Laboratories, 08/2018.

18. REVISION HISTORY

Version	Date	Section	Reason	Reviser	Approval
1	9/2/20	4.2	Added tech code to flex labeling	L Barrett	R SanLuis
1	9/2/20	6.3	Added QC required after every new flex	L Barrett	R SanLuis
1	9/2/20	16	Added QC handoff log	L Barrett	R SanLuis

19. ADDENDA

None