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:

B-type Natriuretic Peptide (BNP) by ADVIA Centaur CP SGAH.C73 v8

Description of change(s):

SOP revised to match current process (*new QC material is already in use*)

Section	Reason
6.1, 6.2	Update QC material and storage, Removed references to Vista analyzer
7.2	Specify freezer requirements by product
7.3	Add pour off tubes
17	Update QC product

This revised SOP 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	**B-type Natriuretic Peptide (BNP) by ADVIA Centaur CP**
Prepared by	Ashkan Chini Date: 5/1/2012
Owner	Robert SanLuis Date: 6/20/2013

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

TABLE OF CONTENTS

1. TEST INFORMATION

Assay	Method/Instrument	Local Code
B-type Natriuretic Peptide	ADVIA Centaur CP	BNPT

Synonyms/Abbreviations
BNP

Department
Chemistry

Form revised 2/02/2007

2. ANALYTICAL PRINCIPLE

The ADVIA Centaur BNP assay is a fully automated two-site sandwich immunoassay using direct chemiluminescent technology, which uses constant amounts of two monoclonal antibodies. The first antibody, in the Lite Reagent, is an acridinium ester labeled monoclonal mouse anti-human BNP F(ab') fragment specific to the ring structure of BNP. The second antibody, in the Solid Phase, is a biotinylated monoclonal mouse anti-human antibody specific to the C-terminal portion of BNP, which is coupled to streptavidin magnetic particles.

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 plasma 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 (K2 EDTA) None
Collection Container	Lavender Top Tube
Volume - Optimum **- Minimum**	Full Tube 1 mL
Transport Container and Temperature	Collection container or Plastic vial at room temperature, **as BNP is unstable in glass containers.**
Stability & Storage Requirements	Room Temperature: 4 hours
	Refrigerated: 2-8° C 24 hours
	Frozen: N/A
Timing Considerations	If unable to run the test within 4 hours, then centrifuge the sample and store separated plasma at 2 - 8° C until testing.
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.

Criteria	
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	Ensure that the samples are free of fibrin or other particulate matter. Samples need to be free of bubbles. **Note:** Use of transfer pipettes affects accurate quantitation of BNP.

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.

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 / Kits	Supplier & Catalog Number
ADVIA Centaur BNP	Siemens reagent cartridge Cat. No. 02816138
ADVIA Centaur R1 Acid Reagent	Siemens reagent Cat. No. 00497043
ADVIA Centaur R2 Base Reagent	Siemens reagent Cat. No. 00497043
ADVIA Centaur Wash 1 Solution	Siemens reagent Cat. No. 01137199
ADVIA Centaur CSC	Siemens reagent Cat. No. 09908593

4.2 Reagent Preparation and Storage

Assay Kit: ADVIA Centaur BNP Ready Pack (primary reagent pack)	
Reagent a	Lite Reagent (Volume 10.5 mL)
Reagent b	Solid Phase (Volume 21 mL)
Manufacturer's Information	Centaur CP BNP Master Curve Cards
Storage	Store the reagent up right at 2-8°C
Stability	Reagent is stable until expiration date stamped on the pack label. Onboard reagents are stable for 41.6 days.
Preparation	Mix all primary reagent packs by hand before loading them onto the system. Visually inspect the bottom of the reagent pack to ensure that all particles are dispersed and re-suspended.

Reagent	**ADVIA Centaur R1 Acid Reagent & R2 Base Reagent**
Container	Reagent bottle (300 mL)
Storage	Store at 2-25°C. Use at 18-30°C.
Stability	Reagent is stable until expiration date stamped on the pack label. Onboard reagents are stable for 30 days. Avoid exposure to light.
Preparation	This reagent is liquid and ready to use.

Reagent	**ADVIA Centaur Wash 1 Solution**
Container	Reagent bottle (1500 mL)
Storage	Store at 2-25°C
Stability	Reagent is stable until expiration date stamped on the pack label. Onboard reagents are stable for 30 days.
Preparation	This reagent is liquid and ready to use.

Reagent	**ADVIA Centaur Cleaning Solution Concentrate**
Container	Reagent bottle (70 mL)
Storage	Store at 2-8°C
Stability	Reagent is stable until expiration date stamped on the pack label. Prepared reagent is stable for 7 days.
Preparation	1. Carefully pour a container of ADVIA Centaur CP Cleaning Solution Concentrate into the cleaning bottle. 2. Add enough reagent grade water to the bottle or container to bring the total volume of cleaning solution to 2 liters.

5. CALIBRATORS/STANDARDS

5.1 Calibrators/Standards Used

Calibrator	Supplier and Catalog Number
Low and High Calibrator	Siemens BNP calibrator, Cat. No. 02817266

5.2 Calibrator Preparation and Storage

Calibrator	**BNP Calibrator (Low & High)**
Preparation	• Add 2.0 mL of reagent grade water into each calibrator vial. • Let the calibrators stand for 15 to 20 minutes at room temperature (20-30°C) to allow the lyophilized material to dissolve. • Gently swirl and invert the vials until homogeneous.

Storage/Stability	• Store at 2-8°C • **Unopened Calibrator** is stable until the expiration date on the vial. • **Opened Calibrator:** once reconstituted use immediately and freeze the rest. The frozen calibrator is stable for 60 days when stored at ≤ -20°C. **Freeze-thaw is recommended only one time after reconstitution.**

5.3 Calibration Procedure

Criteria	Special Notations
Frequency	• When the calibration interval expires • Calibration Interval is 28 days • A calibration is invalid • New lot or shipment of assay reagents • Controls are repeatedly out of range.

	Calibration Procedure
1.	From the main page, go to **Result** > **Pending**, and make sure nothing is pending for this method. **Note**: The instrument must be in a **Ready** mode before moving on to the next step.
2.	Get the **Centaur CP** Calibrator Master Curve card from the calibrator pack and reagent Master Curve card from the reagent box. From the main page, go to **Definition** > **Calibrators** > **Scan**, scan all 3 barcodes (on the Calibrator Master Curve) from top to bottom. Then go to **Definition** > **Master Curves** > **Scan**, scan all barcodes (on the Reagent Master Curve) from top to bottom. **Notes**: • The calibrator pack and reagent box include both Centaur CP and XP master curves. Be sure to scan the correct master curve. • Always scan the calibrator information first.
3.	Load the low and high calibrators into appropriate sample pour-off tubes that accommodate the Siemens-supplied barcode label. **Note**: The low and high calibrators provided in this kit are matched to the original primary reagent pack. Do not mix calibrator lots with different lot of reagent packs.
4.	Attach the Siemens-supplied barcode labels to the pour off tubes.
5.	Load the sample tubes into a sample rack. Move the tube type selector on the rack to position A.
6.	Load the rack in the sample compartment.
7.	On the Sample Compartment screen, select the lane containing the sample rack with the calibration material.
8.	Confirm or enter the calibrator information.

	Calibration Procedure
9.	Select **Close** to return to the workspace.
10.	At the workspace, select the primary reagent area.
11.	Select the assay to calibrate.
12.	Select **Calibrate**. The system automatically begins sampling the calibrator material. **NOTE:** The Calibrate button is not active on the Primary Reagent Screen in the following situations: • there are no calibrators defined • there are no Master Curves defined • there is no reagent onboard • the calibration material for defined lots is expired • there is no calibrator material in the sample compartment
13.	Select **Close** to return to the workspace.
14.	To check the time due for the calibration: a. At the workspace, select **Results**. b. Select the Calibrations tab to view the Time Due stamp for the assay you are calibrating. c. Select **Close**.

5.4 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™ Cardiac Markers Plus Control LT Levels 1C, 2 and 3	Bio-Rad Laboratories Cat # 297, 298 and 299

6.2 Control Preparation and Storage

Control	Liquichek Cardiac Markers Plus Control LT, Level 1C, 2 and 3
Preparation	Allow the frozen control to thaw at room temperature (18-25°C) for approximately 30 minutes or until completely thawed. Swirl

	the contents gently to ensure homogeneity. (Do not use a mechanical mixer. Transfer QC into the sample pour off tube, load in appropriate rack then run QC. 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 stored unopened at 2-8°C, stable for 10 days. Thawed and opened: When stored open at 2- 8°C, stable for 10 days. Once thawed, do not re-freeze

6.3 Frequency

Analyze all 3 levels of QC material after each calibration.
QC is run daily as follows: Levels 1 and 3 are run on day shift, Level 2 on evening shift, and either Level 1 or 3 on night shift.

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.

IF the Quality Control …	THEN…
does not fall within the Expected Values	• Verify that the materials are not expired. • Verify that required maintenance was performed. • Verify that the assay was performed according to the instructions for use. • Rerun the assay with fresh quality control samples. • If necessary recalibrate the assay and repeat quality control • If unable to resolve issue and/or instrument is inoperable, testing may be performed by alternative method B-type Natriuretic Peptide (BNP) by Triage Meter. Refer to the appropriate SOP

NOTE: The laboratory director or designee may override rejection of partial or complete runs. Justification for the override must be documented in detail.

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

ADVIA Centaur CP Immunoassay 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

- Purified water (Millipore® or equivalent)
- Calibrated pipettes and disposable tips
- Cuvettes
- Tips
- Sample pour off tubes

8. PROCEDURE

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	Loading samples into Sample Rack
1.	Position the barcode label vertically on the sample tube approximately 2 cm (0.8 in) from the top.
2.	Move the tube-type selector on the rack to position B for short and C for long sample tubes.
3.	Place the sample tubes in the rack.
4.	Ensure that there is only one type of sample tube in the rack and that the tube-type selector is positioned correctly for that tube type. Ensure that the barcode labels are clearly visible above or between the slots in the rack.

8.2	Loading Sample Racks into the System
1.	Open the sample compartment door.
2.	Slide the sample rack into the appropriate lane. Use one continuous motion. The sample barcodes are read as the rack is inserted into the system. If the rack stops, or reverses, during its insertion, the barcodes may not be read.
3.	Ensure that the sample rack is inserted correctly, and locked into place. When the sample rack is inserted correctly, you will hear and feel it click into place.
4.	Close the sample compartment door.

8.3	Scheduling Samples through the Sample Compartment Screen
1.	At the workspace, select the sample compartment. Select the Sample Rack. Select a sample
2.	Select the sample type by toggling the sample selection button. The default is a routine patient sample (**smp**). If you want the sample to be processed before routine samples, select **stat**.
3.	Select the Sample ID field. The samples are numbered 1-12. Number 1 is the sample at the back of the sample compartment, and number 12 is the sample at the front of the sample compartment.
4.	If the sample ID was not entered using a barcode label on the sample tube, enter the sample ID and then press **Enter**.
5.	Select the assays or the profiles to process against the sample.
6.	Select **Close**.

9. CALCULATIONS

The instrument automatically calculates the concentration of BNP in pg/mL.

10. REPORTING RESULTS AND REPEAT CRITERIA

10.1 Interpretation of Data

None required

10.2 Rounding

No rounding is necessary. Instrument reports results in whole numbers.

10.3 Units of Measure

pg/mL

10.4 Clinically Reportable Range (CRR)

5 - 5,000 pg/mL

10.5 Review Patient Data

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

10.6 Repeat Criteria and Resulting

IF the result is …	THEN…
< 5 pg/mL	Assure there is sufficient sample devoid of bubbles, cellular debris, and/or fibrin clots. Repeat test. Report as: < 5 pg/mL-REP
≥ 5000 pg/mL	Report as ≥ 5000 pg/mL

To manually enter results in the LIS, use worksheet code SCH1 (at SGAH) or WHE1 (at WAH) with the Centaur default method code CS1 (at SGAH) or CW1 (at WAH).

11. EXPECTED VALUES

11.1 Reference Ranges

0 – 100 pg/mL

11.2 Critical Values

None established

11.3 Standard Required Messages

None established

12. CLINICAL SIGNIFICANCE

This assay is indicated for the measurement of plasma BNP as an aid in the diagnosis and assessment of the severity of heart failure. In patients with acute coronary syndromes (ACS), this test, in conjunction with other known risk factors, can also be used to predict survival as well as to predict the likelihood of future heart failure.

Heart failure is an important clinical syndrome which compromises left ventricular systolic or diastolic function or a combination of both. Heart failure occurs when the heart is unable to pump blood at a rate sufficient for metabolic requirements. Its most common causes are coronary artery disease, hypertension, valvular heart diseases and cardiomyopathies. Accurate and early diagnosis is important since effective therapeutic interventions (e.g., angiotensin converting enzyme inhibitors, beta-blockers) are available, which improve both morbidity and mortality. Based on clinical signs and symptoms, the severity of heart failure

is classified into four classes of increasing disease progression according to the New York Heart Association classification.

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 containing such error messages should be held for follow-up. Refer to the ADVIA Centaur CP Operator's Guide.

14. LIMITATIONS OF METHOD

14.1 Analytical Measurement Range (AMR)

2 - 5000 pg/mL

14.2 Precision

Six samples were assayed 4 times, in 20 runs, on 2 systems over a period of 20 days. The following results were obtained:

Mean pg/mL	Within-Run % CV	Run-to-Run % CV	Total CV %
43	2.7	3.3	4.2
455	2.2	3.3	3.9
1771	1.9	2.9	3.5

14.3 Interfering Substances

Heterophilic antibodies in human serum can react with reagent immunoglobulins, interfering with in vitro immunoassays. Patients routinely exposed to animals or to animal serum products can be prone to this interference and anomalous values may be observed. Additional information may be required for diagnosis.

Specimens that are….	Demonstrate ≤ 10 % change in results up to…
Lipemic	800 mg/dL of triglycerides 1000 mg/dL of cholesterol
Uremic	200 mg/dL of urea 2.5 mg/dL of creatinine
Icteric	25 mg/mL of unconjugated bilirubin
Hemolyzed	100 mg/dL of hemoglobin

14.4 Clinical Sensitivity/Specificity/Predictive Values

N/A

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.

ADVIA Centaur BNP Ready Pack contains sodium azide which can react with copper and lead plumbing to form explosive metal azides.

16. RELATED DOCUMENTS

1. ADVIA Centaur CP Operator's Manual
2. Laboratory Quality Control Program
3. QC Schedule for ADVIA Centaur CP
4. Laboratory Safety Manual
5. Safety Data Sheets (SDS)
6. Quest Diagnostics Records Management Procedure
7. Centrifuge Use, Maintenance and Functions Checks (Lab policy)
8. Hemolysis, Icteria and Lipemia Interference (Lab policy)
9. Repeat Testing Requirement (Lab policy)
10. ADVIA Centaur CP Sample Processing, Startup and Maintenance (Chemistry SOP)
11. Current Allowable Total Error Specifications
    at http://questnet1.qdx.com/Business_Groups/Medical/qc/docs/qc_bpt_tea.xls
12. Current package insert BNP Reagent Cartridge

17. REFERENCES

1. Package Insert, Liquichek Cardiac Markers Plus Control LT, Bio-Rad Laboratories, 12/2015.
2. Package Insert, BNP Calibrator, Siemens Diagnostics revised 09/2014
3. Package Insert, BNP reagent pack, Siemens Diagnostics revised 06/2015

18. REVISION HISTORY

Version	Date	Section	Reason	Reviser	Approval
000	6/1/12	3.2	Remove centrifugation requirement	J.Buss	J.Buss, RSL
000	6/1/12	4.1, 4.2	Remove Multi-Diluent 1 (not used)	J.Buss	J.Buss, RSL
000	6/1/12	10.4	Edit CRR at Medical Director request	J.Buss	J.Buss, RSL
000	6/1/12	10.5	Match CRR; No dilutions performed. Add LIS worksheet & method codes	J.Buss	J.Buss, RSL
000	6/1/12	14.1	Edit lower AMR	J.Buss	J.Buss, RSL
001	6/20/13		Update owner	L Barrett	R SanLuis
001	6/20/13	5.2	Remove open storage at frozen temp	A Chini	R SanLuis
001	6/20/13	5.3	Shorten calibrator interval to 28 days	A Chini	R SanLuis
001	6/20/13	6.4	Removed GEC SOP as alternate method	A Chini	R SanLuis

001	6/20/13	14.3	Revise % change and add hemolyzed	A Chini	R SanLuis
001	6/20/13	15	Added specific safety notes	A Chini	R SanLuis
001	6/20/13	16	Added Centaur operation SOP	L Barrett	R SanLuis
002	4/10/15	4.1	Update catalog numbers	A Chini	R SanLuis
002	4/10/15	6.2	Change open storage to 8 days	A Chini	R SanLuis
002	4/10/15	6.4, 6.6	Replace LIS with Unity Real Time	A Chini	R SanLuis
002	4/10/15	14.1	Change lower limit of AMR to match PI	A Chini	R SanLuis
002	4/10/15	14.2	Change data to match update PI	A Chini	R SanLuis
002	4/10/15	Footer	Version # leading zero's dropped due to new EDCS in use as of 10/7/13	L Barrett	R SanLuis
3	7/7/15	6.3	Change frequency to daily to match log	L Barrett	R SanLuis
4	3/15/16	1	Update local code	A Chini	R SanLuis
4	3/15/16	3.2	Change room temp. stability to 4 hours, add note for transfer pipettes	A Chini	R SanLuis
4	3/15/16	4.2	Add hazard information	A Chini	R SanLuis
4	3/15/16	5.3	Update steps 1 - 3	A Chini	R SanLuis
5	10/5/16	Header	Add WAH	L Barrett	R SanLuis
5	10/5/16	4,5,6	Remove individual section labeling instructions and add general one	L Barrett	R SanLuis
5	10/5/16	10.5	Move patient review from section 6	L Barrett	R SanLuis
5	10/5/16	10.6	Add repeat test if <5	L Barrett	R SanLuis
5	10/5/16	15	Update to new standard wording, add reagent warning from section 4, remove liquid waste disposal.	L Barrett	R SanLuis
6	1/23/17	6.1, 6.2	Update QC material and storage	L Barrett	R SanLuis
6	1/23/17	7.2	Change freezer temp from -70 to -50	L Barrett	R SanLuis
6	1/23/17	17	Update QC product	L Barrett	R SanLuis
7	2/16/17	6.2	Removed references to Vista analyzer	J Negado	R SanLuis
7	2/16/17	7.3	Add pour off tubes	J. Negado	R. SanLuis

19. ADDENDA

None