DXI (BNPEP) B-TYPE NATRIURETIC PEPTIDE

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PURPOSE

To provide instructions for how to perform BNPEP testing on the DXI instruments**.**

PRINCIPLE

The Alere Triage BNPEP reagent, when used in conjunction with the Beckman DXI Systems and Alere Triage Calibrators, is intended for quantitative determination of BNPEP concentration in human plasma.

BACKGROUND

Clinical Significance

It is estimated that 5.8 million people in the United States have heart failure, with approximately 670,000 new cases occurring each year. Congestive heart failure (CHF) occurs when the heart cannot deliver a sufficient amount of blood to the body. This condition can occur at any age but is most prevalent in an aged population. Symptoms of CHF include shortness of breath, fluid retention and respiratory distress. These symptoms are often vague and nonspecific for detecting early stages of CHF.

B-type natriuretic peptide (BNP) is a member of a class of hormones that regulate blood pressure. The heart is the main source of circulating BNP in humans. The molecule is released into the blood in response to increased heart pressure. Various studies have demonstrated that increased levels of circulating BNP are found in early stages of CHF. The level of BNP in the blood continues to increase as the CHF disease advances. The Triage BNP test offers as objective, noninvasive measurement for assessing patients for CHF and risk stratification in patients with acute coronary syndromes (ACS).

Methodology

The Triage BNP test is a two-site immunoenzymatic (“sandwich”) assay. A sample is added to a reaction vessel with mouse monoclonal anti-human BNP antibody-alkaline phosphatase conjugate and paramagnetic particles coated with mouse omniclonal anti-human BNP antibody. BNP in human plasma binds to the immobilized anti-BNP on the solid phase, while the mouse anti-BNP conjugate reacts specifically with bound BNP. After incubation in a reaction vessel, materials bound to the solid phase are held in a magnetic field while unbound materials are washed away. A chemiluminescent substrate, Lumi-Phos 530, is added to the reaction vessel and the light generated by the reaction is measured with a luminometer. The light production is directly proportional to the concentration of BNP in the sample. The amount of analyte in the sample is determined from a stored, multi-point calibration curve.

RELATED DOCUMENTS

R-PO-CH-0810	Quality Control Program General Laboratory
R-PO-CH-0809	Quality Control Westgard Rules Statistics
R-PR-AD-0540	Specimen Rejection/Cancellation Protocol
J-F-CG-0824	DXI & Access Controls
J-F-CH-0825	DXI Calibrators
M-F-CH-0820	Chemistry Controls
M-F-CH-0826	Chemistry Calibrators
R-F-CH-2000	DXI Analytical Measurement Range

SPECIMEN

Type of Specimen

Biological fluid samples should be collected in the same manner routinely used for any laboratory test. Freshly drawn plasma is the preferred specimen.

Specimen Storage and Stability

- Plasma samples in plastic or glass tubes 24 hours refrigerated or 8 hours at room temperature

Criteria for Unacceptable Specimens

Refer to the PROCEDURAL NOTES section of this chemistry information sheet for information on unacceptable specimens.

SAMPLE VOLUME

A filled 0.5 mL sample cup is the optimum volume. For optimum primary sample tube volumes in primary tube samples and minimum volumes, refer to the Primary Tube Sample Template for your system.

REAGENTS

Triage® BNP Reagent Pack
Cat. No. 98200: 100 determinations, 2 packs, 50 tests/pack •
Provided ready to use.

- Store upright and refrigerate at 2 to 10 °C.
- Refrigerate at 2 to 10 °C for a minimum of two hours before use on the instrument.
- Stable until the expiration date stated on the label when stored at 2 to 10 °C.
- Stable at 2 to 10 °C for 28 days after initial use.
- Signs of possible deterioration are a broken elastomeric layer on the pack or control values out of range.
- If the reagent pack is damaged (i.e., broken elastomer), discard the pack.
- All antisera are polyclonal unless otherwise indicated.

R1a: Paramagnetic particles coated with mouse omniclonal anti-human BNP antibody suspended in TRIS buffered saline, with bovine serum albumin (BSA), 0.1% ProClin 300, and < 0.1% sodium azide.

R1b: Purified mouse and goat IgG in TRIS buffered saline with 0.1% ProClin 300 and < 0.1% sodium azide.

R1c: Mouse monoclonal anti-human BNP antibody-alkaline phosphatase bovine conjugate in PBS buffered saline with BSA, 0.1% ProClin 300, and < 0.1% sodium azide.

- For *In Vitro* Diagnostic Use.
- Patient samples and blood-derived products may be routinely processed with minimum risk using the procedure described. However, handle these products as potentially infectious according to universal precautions and good clinical laboratory practices, regardless of their origin, treatment, or prior certification. Use an appropriate disinfectant for decontamination. Store and dispose of these materials and their containers in accordance with local, state and federal regulations and guidelines.
- Sodium azide may react with lead and copper plumbing to form highly explosive metal azides. On disposal of liquids, flush with a large volume of water to prevent azide build-up.
- The Triage® BNP test should not be used as absolute evidence for CHF. The results should be interpreted along with clinical findings and other laboratory test results.
- Blood concentrations of BNP may be elevated in patients who are experiencing a heart attack, patients that are candidates for renal dialysis, and patients that have had renal dialysis.
- ProClin 300 is a potential skin sensitizer. Avoid spilling or splashing this reagent on skin or clothing. In case of contact with the reagent, flush thoroughly with soap and water.
- The Material Safety Data Sheet (MSDS) is available upon request.

R1 Triage® BNP Reagent Packs

1. **Triage® BNP Calibrators**
   Provided at zero, and approximately 25, 100, 500, 2500, and 5000 pg/mL. Cat. No. 98202
   Storage for calibrators is at -20 degrees C or colder. Thaw only once. Stable for 30 days at 2-8 degrees C after thawing.

2. **Triage® BNP QC Controls or other commercially available control material.**
   Provided at approximately 80, 400, and 2200 pg/mL. Cat. No. 98201
   Storage for calibrators is at -20 degrees C or colder. Thaw only once. Stable for 30 days at 2-8 degrees C after thawing.

3. **Access Substrate** Cat. No. 81906

4. **DXI Wash Buffer**
   NOTE: The required wash buffer catalog number is dependant upon your current instrument status. Please contact Beckman Coulter technical support if you are unsure of which buffer to order.
   Cat. No. 81907 (Access, Access 2, SYNCHRON LXi, UniCel DxC600i) Cat. No. 8547197 (UniCel DxI) Or
   Cat. No. A16792 (Access, Access 2, SYNCHRON LXi, UniCel DxC600i)
   Cat. No. A16793 (UniCel DxX 660i, UniCel DxC 680i, UniCel DxC 860i, UniCel DxC 880i, UniCel DxI 600, UniCel DxI 800)

5. **One of the following immunoassay systems:**
   Access, Access 2, Synchron LXi 725, UniCel DxC 660i, UniCel DxC 680i, UniCel DxC 860i, UniCel DxC 880i, UniCel DxI 600, UniCel DxI 800 or UniCel DxC600i

CALIBRATION

An active calibration curve is required for all tests. For the Triage BNP assay, calibration is required every 28 days. Refer to the appropriate system manuals and/or Help system for information on calibration theory, configuring calibrators, calibrator test request entry, and reviewing calibration data.

The Triage BNP Calibrators are provided at six levels - zero and approximately 25, 100, 500, 2500 and 5000 pg/mL. Assay calibration data are valid up to 28 days.

Calibrators run in duplicate.

QUALITY CONTROL

See Related Documents

PROCEDURE STEPS

1. Instrument: Refer to the appropriate system manuals and/or Help system for preparation and operation.

2. Assay Procedure: Refer to the appropriate system manuals and/or Help system for information on managing samples, configuring tests, requesting tests, and reviewing test results.

RESULTS

Patient test results are determined automatically by the system software using a smoothing spline math model. The amount of analyte in the sample is determined from the measured light production by means of the stored calibration data. Patient test results can be reviewed using the appropriate screen. Refer to the appropriate system manuals and/or Help system for complete instructions on reviewing sample results.

PERFORMANCE CHARACTERISTICS

Reference ranges

Reference Range	Interpretation
<100 pg/mL	Negative
≥ 100 pg/mL	Positive
> 200 pg/mL	High Correlation with left ventricular heart failure
"Patients with myocardial infarction and patients who are candidates for renal dialysis, or have undergone renal dialysis, may have elevated BNP levels."	

Analytic Measurement Range (AMR)

Samples can be accurately measured within the analytic range of the lower limit of detection and the highest calibrator value (approximately 5–5000 pg/mL).

- If a sample contains less than the lower limit of detection for the assay, report the results as less than that value (i.e. < 5 pg/mL).
- If a sample contains more than the stated value of the highest Triage BNP Calibrator (S5)(>5000 pg/mL), report as >5000 pg/mL. **DO NOT DILUTE.**

LIMITATIONS

Substances

Hemoglobin (up to 500 mg/dL), triglycerides (triolein up to 3000 mg/dL), bilirubin (conjugated up to 20 mg/dL), fibrinogen (up to 800 mg/dL) or human serum albumin (up to 1500 mg/dL) added to plasma specimens containing BNP did not interfere with the recovery of BNP.

Analytical Specificity

Pharmaceuticals

The following drugs were evaluated for potential cross-reactivity and interference in the Triage BNP test. All drugs were tested at concentrations representing the blood concentrations that would result from a maximal therapeutic dose and at least twice the maximal therapeutic dose. None of the drugs interfered with the recovery of BNP. Additionally, these drugs did not produce a significant response when tested in a specimen not containing BNP. There was no significant interference with the BNP measurement, nor was there any assay cross reactivity.

Acetaminophen	Aspirin	Cocaine	Furosemide	Nitrofurantoin	Quinidine
Allopurinol	Atenolol	Diclofenac	Heparin	Nystatin	Theophylline
Ambroxol	Caffeine	Digoxin	Ibuprofen	Oxytetracycline	Trimethoprim
Ampicillin	Captopril	Dopamine	Methyldopa	Phenytoin	Verapamil
Ascorbic Acid	Cinnarizine	Erythromycin	Nifedipine	Propranolol	

Nesiritide is a synthetic form of BNP; BNP measurements should not be performed during Nesiritide Infusion.

Proteins and Peptides

The following proteins and peptides were evaluated for potential cross reactivity and interference in the Triage® BNP test at the concentrations indicated below. There was no significant interference with the BNP measurement, nor was there any significant assay cross-reactivity.

Reactivity with Related Proteins and Peptides

Substance	Concentration of Substance	% Recovery
Adrenomedullin	1000 pg/mL	101.4%
α-Atrial Natriuretic polypeptide 1-28	1000 pg/mL	99.2%
Angiotensin I	600 pg/mL	97.9%
Angiotensin II	600 pg/mL	96.5%
Angiotensin III	1000 pg/mL	95.1%
Arg Vasopressin	1000 pg/mL	95.7%
C type Natriuretic Peptide 53	1000 pg/mL	96.8%
Endothelin I	20 pg/mL	99.2%
Prepro ANF 104-123	1000 pg/mL	96.7%
Prepro ANF 26-55	1000 pg/mL	94.2%
Prepro ANF 1-21	1000 pg/mL	98.5%
Prepro ANF 22-46	1000 pg/mL	97.7%
Renin	50 ng/mL	95.8%
Urodilantin	1000 pg/mL	91.6%

Substance	Level Tested	Effect
Bilirubin	20 mg/dL	No significant interference
Hemoglobin	500 mg/dL	No significant interference
Triglycerides	3000mg/dL	No significant interference

PROCEDURAL NOTES

1. Refer to the appropriate system manuals and/or Help system for a specific description of installation, startup, principles of operation, system performance characteristics, operating instructions, calibration procedures, operational limitations and precautions, hazards, maintenance, and troubleshooting.

2. Mix contents of new (unpunctured) reagent packs by gently inverting pack several times before loading on the instrument. Do not invert open (punctured) packs.

3. The Triage BNP test does not demonstrate any “hook” effect up to BNP concentrations greater than 500,000 pg/mL

4. The Triage BNP test results should be interpreted in light of the total clinical presentation of the patient, including: clinical history, data from additional tests and other appropriate information.

REFERENCES

Alere* Access BNP assay procedure version 26259en Rev. D, 09/2016

BNP Sample Stability Study, University of Maryland, 2004