

**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
DEVICE ONLY TEMPLATE**

A. 510(k) Number:

k043476

B. Purpose for Submission:

Addition of NT-proBNP assay to the Stratus® CS STAT Fluorometric Analyzer

C. Analyte:

B-type natriuretic peptide test system (BNP)

D. Type of Test:

Quantitative

E. Applicant

Dade Behring, Inc.

F. Proprietary and Established Names:

Stratus CS® Acute Care™ NT-proBNP (pBNP) TestPak assay

Stratus CS® pBNP CalPak

Stratus CS® pBNP DilPak

G. Regulatory Information:

1. Regulation section:
21 CFR 862.1117, B-type natriuretic peptide test system
21 CFR 862.1150, calibrator secondary
2. Classification:
Class II
3. Product Code:
NBC; JIT
4. Panel:
Chemistry (75)

H. Intended Use:

1. Intended use(s):
The Stratus CS® Acute Care™ NT-proBNP (pBNP) is an *in vitro* diagnostic test for the quantitative determination of N-terminal pro-brain natriuretic peptide (NT-proBNP) in heparinized plasma. In individuals suspected of having congestive heart failure (CHF), measurements of NT-proBNP are used as an aid in the diagnosis and assessment of severity. The test is further indicated for the risk stratification of patients with acute coronary syndrome and heart failure.

2. Indication(s) for use:

The Stratus® CS Acute Care™ NT-proBNP method (pBNP) is an *in vitro* diagnostic assay for the quantitative determination of N-terminal pro-brain natriuretic peptide (NT-proBNP) in human plasma. In individuals suspected of having congestive heart failure (CHF), measurements of NT-proBNP are used as an aid in the diagnosis and assessment of severity. The test is further indicated for the risk stratification of patients with acute coronary syndrome and heart failure.

3. Special condition for use statement(s):

Prescription use

4. Special Instrument Requirements:

Stratus® CS STAT Fluorometric Analyzer

I. Device Description:

The Stratus® CS Acute Care™ pBNP TestPak is required to perform the pBNP test. The TestPak contains liquid reagents. Each box contains 100 TestPaks. Active ingredients are: Alkaline phosphatase conjugated to anti-NT-proBNP (source sheep polyclonal), dendrimer linked NT-proBNP antibody (sheep polyclonal), and 4-methylumbelliferyl phosphate.

The Stratus CS® pBNP CalPak is a frozen liquid product containing synthetic human NT-proBNP in a bovine albumin matrix with stabilizers and preservative. The kit consists of five CalPaks at a single calibrator level. Each CalPak contains calibrator reagent in three wells.

The Stratus CS® pBNP DilPak is a refrigerated product containing a buffered bovine protein matrix with stabilizers and preservative. The kit consists of 5 DilPaks with diluent in one well.

J. Substantial Equivalence Information:

1. Predicate device name(s):

Roche Diagnostics Elecsys® proBNP Immunoassay

2. Predicate K number(s):

K022516

3. Comparison with predicate:

Similarities		
Item	Stratus CS® Acute Care™ NT-proBN	Roche NT-proBNP
Assay type	Immunoassay (fluorometric)	Immunoassay (electrochemiluminescent)
Antibody	Polyclonal sheep antibody	Polyclonal sheep antibody
Cut-off	125 pg/mL for patients <75 years 450 pg/mL for patients ≥ 75 years	125 pg/mL for patients <75 years 450 pg/mL for patients ≥ 75 years
Reference	Roche purified synthetic NT-proBNP	Roche purified synthetic NT-proBNP
Differences		
Item	Stratus CS® Acute Care™ NT-proBN	Roche NT-proBNP
Indications for Use	For the <i>in vitro</i> quantitative determination of N-terminal pro-brain natriuretic peptide in human plasma as an aid in the diagnosis and assessment of severity of individuals suspected of having congestive heart failure. The test is further indicated for the risk stratification of patients with acute coronary syndrome and heart failure.	<i>in vitro</i> quantitative determination of NT-proBNP in human serum and plasma as an aid in the diagnosis of individuals suspected of having congestive heart failure. The test is further indicated for the risk stratification of patients with acute coronary syndrome and congestive heart failure.
Reportable range	15-20,000 pg/mL	5-35,000 pg/mL
Analytical sensitivity	15 pg/mL	5 pg/ml
Sample volume	50 µL	20 µL

K. Standard/Guidance Document Referenced (if applicable):

NCCLS EP 5-A, 1999; Class II Special Controls Guidance Document for B-Type Natriuretic Peptide Premarket Notifications: Final Guidance for Industry and FDA Reviewers (11/30/2000).

L. Test Principle:

The Stratus CS® Acute Care™ NT-proBNP (pBNP) method is a two-site sandwich assay based upon solid phase Radial Partition Immunoassay (RPIA) technology. Dendrimer linked polyclonal antibody is added to the center portion of a square piece of glass fiber paper in the pBNP TestPak. This antibody recognizes a distinct antigenic site on the NT-proBNP molecule. Sample is then added onto the paper where it reacts with the immobilized antibody. After a short incubation, a conjugate consisting of enzyme-labeled polyclonal antibody directed against a second distinct

antigenic site on the NT-proBNP molecule is pipetted onto the reaction zone of the paper. During this second incubation period, enzyme-labeled antibody reacts with the bound NT-proBNP, forming an antibody-antigen-labeled antibody sandwich. The unbound labeled antibody is later eluted from the field of view of the Stratus® CS analyzer by applying a substrate wash solution to the center of the reaction zone. By including substrate for the enzyme within the wash solution, initiation of enzyme activity occurs simultaneously with the wash. The enzymatic rate of the bound fraction increases directly with the concentration of NT-proBNP in the sample. The reaction rate can then be measured by an optical system that monitors the reaction rate via front surface fluorescence. All data analysis functions are performed by the microprocessor within the analyzer.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Sample	Mean (pg/mL)	Within-Run Precision		Total Precision	
		SD (pg/mL)	% CV	SD (pg/mL)	% CV
Human plasma pool 1	153	6.5	4.2	7.8	5.1
Human plasma pool 2	463	15.0	3.2	15.0	3.2
Human plasma pool 3	6726	177.3	2.6	177.3	2.6
Control Level 1	156	7.7	4.9	7.7	4.9
Control Level 2	5371	115.5	2.1	158.6	3.0

Precision testing was done in accordance with NCCLS EP5-A, 1999. Specimens at each level were analyzed in duplicate once per day for 20 days.

b. Linearity/assay reportable range:

The reportable range of the assay is from 15-20,000 pg/mL. Two high pBNP plasma samples (pBNP = 19,260 pg/mL and 21,453 pg/mL) were diluted across the expected range with a low pBNP sample to produce 5 levels. High range linearity was evaluated by comparing observed vs. expected values obtained with the pBNP method. A linear regression analysis was then performed on the data to yield the following: slopes = 1.001 and 0.995 respectively, $r = 0.999$ for both samples, intercept = 61.8 pg/mL and -129.4 pg/mL respectively. Lower range linearity was evaluated by diluting a patient sample (pBNP = 1231 pg/mL) with a low pBNP sample across the assay range to produce 5 levels. A linear regression analysis was performed on the data to yield the following: slope = 1.00, $r = 0.999$, intercept = -4.7.

Hook effect was evaluated using samples containing NT-proBNP concentrations ranging from 0 to 1,400,000 pg/mL. Data indicated no hook effect up to 1,400,000 pg/mL.

c. *Traceability (controls, calibrators, or method):*

The assay is referenced to Roche purified synthetic NT-proBNP (1-76). The assigned values for the Stratus® CS pBNP Calibrator are referenced to a NT-proBNP master pool. Real time stability was acceptable up to 13 months, to support the 12 month expiration date claim.

d. *Detection limit:*

The analytical sensitivity for the pBNP assay is ≤ 15 pg/mL. This is defined as the concentration at two standard deviations ($n = 20$) of a sample devoid of NT-proBNP. Functional sensitivity was determined by performing a 20 day ANOVA experiment using samples prepared from normal human plasma at appropriate NT-proBNP concentrations. Total % CV was plotted versus the target NT-proBNP concentration for each sample. The functional sensitivity was determined to be ≤ 50 pg/mL.

e. *Analytical specificity:*

No significant interference was found for bilirubin (conjugated) up to 60 mg/dL, bilirubin (unconjugated) up to 60 mg/dL, hemoglobin up to 1000 mg/dL, triglycerides up to 3000 mg/dL, and rheumatoid factor up to 500 IU/mL. The pharmaceutical Natrecor® shows no significant cross reactivity (less than 1 %) at a concentration of 3.5 μ g/mL when added to samples containing 0 and 125 pg/mL NT-proBNP. An extensive list of other compounds was evaluated for interference and was found to have no significant interference or cross reactivity. A list of these compounds is present in the pBNP labeling.

f. *Assay cut-off:*

The recommended medical decision thresholds by age group are:

Patients < 75 years	125 pg/mL
Patients \geq 75 years	450 pg/mL

2. Comparison studies:

a. *Method comparison with predicate device:*

Comparison using split patient heparinized plasma samples between the Dade Behring Stratus® CS Acute Care™ TestPak assay and the predicate Roche Elecsys® proBNP method demonstrated the following method comparison using samples with values ranging from 16.1 – 17,691.9 pg/mL. Samples were obtained from patients known to have CHF ($n = 225$), patients with signs and symptoms suggestive of CHF ($n = 15$) and patients without CHF ($n = 241$).

Comparative Method	Slope	Intercept (pg/mL)	Correlation Coefficient	n
Roche Elecsys® proBNP	0.96	5.5	0.99	481

b. *Matrix comparison:*

Plasma specimens (lithium heparin, sodium heparin) may be used for the pBNP assay. Serum samples should not be used with the pBNP assay. Lithium heparin samples (n = 19) ranging from < 15 to 15,810 pg/mL when compared to sodium heparin samples gave a slope of 1.00, correlation coefficients of 0.999, and intercept of 28 pg/mL using Passing-Bablok regression statistics.

3. Clinical studies:

a. *Clinical sensitivity:*

Clinical Studies: For the Reference Study Group, NT-proBNP concentrations were determined in 308 individuals without congestive heart failure (163 women and 145 men). This population included apparently healthy individuals and individuals with diabetes, hypertension, and pulmonary disease. For the Disease Study Group, NT-proBNP concentrations were determined in 234 patients diagnosed with congestive heart failure (CHF). This population included 70 women and 164 men.

The tables below show the clinical sensitivity and specificity of the Stratus® CS Acute Care™ pBNP assay using a cutoff of 125 pg/mL for patients younger than 75 years and 450 pg/mL for patients 75 years or older.

Males		
	<75 yrs	≥75 yrs
Sensitivity	90% (104/116)	92% (44/48)
95% Confidence Interval	84 - 95	84 - 99
Specificity	92% (76/83)	73% (45/62)
95% Confidence Interval	86 - 98	61 - 84

Females		
	<75 yrs	≥75 yrs
Sensitivity	84% (43/51)	95% (18/19)
95% Confidence Interval	74 - 94	85 - 100
Specificity	92% (95/103)	85% (51/60)
95% Confidence Interval	87 - 97	76 - 94

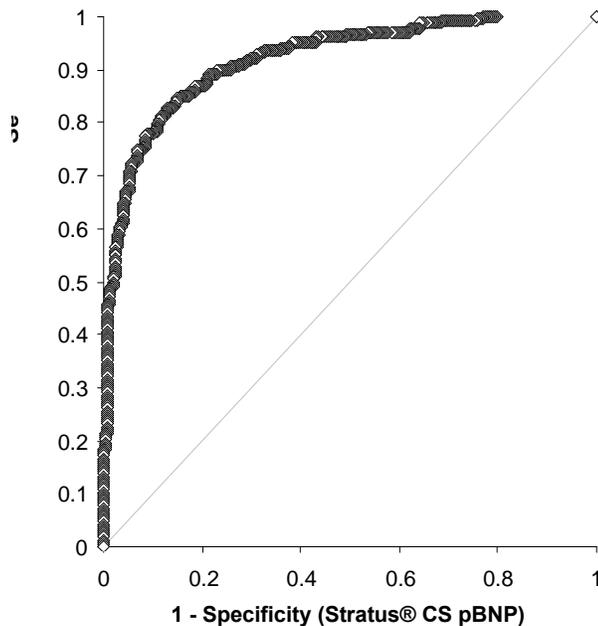
b. *Clinical specificity:*

See Clinical Sensitivity above

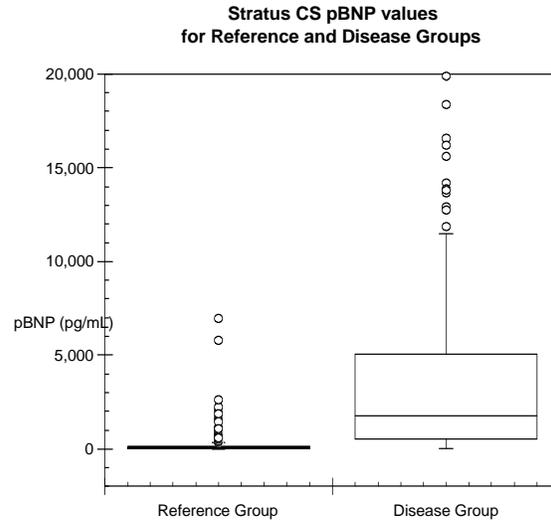
c. *Other clinical supportive data (when a and b are not applicable):*

4. Clinical cut-off:

The Receiver Operator Curve (ROC) presents the clinical sensitivity and specificity at various cutoffs for the 234 patients diagnosed with CHF and 308 subjects without CHF. The ROC curve for the Stratus® CS Acute Care™ pBNP assay is shown below. The area under ROC curve (AUC) is 0.921 with a 95% confidence interval of 0.898 to 0.943. An age-matched ROC analysis of the clinical data was performed via the weighted method described in Kondratovich, M (2002), Matched Receiver Operating Characteristic (ROC) analysis and propensity scores, Proceedings of the 2002 Joint Statistical Meeting, Biopharmaceutical Section, New York, NY. The resulting AUC is 0.924 with a 95% confidence interval of 0.902 to 0.946.



A box and whiskers plot of the clinical study population is presented below. Recommended clinical thresholds are 125 pg/mL for patients younger than 75 years and 450 pg/mL for patients 75 years and older. Nine disease group samples with values above the assay range are not displayed in the plot.



5. Expected values/Reference range:

NT-proBNP concentrations in the Reference Group are shown in the following tables. The recommended medical decision thresholds, by age group, are:

Patients < 75 years: 125 pg/mL

Patients ≥ 75 years: 450 pg/mL

Reference Study Group

NT-proBNP concentrations were determined in 308 individuals without congestive heart failure (163 women and 145 men). This population included apparently health individuals and individuals with diabetes, hypertension, and pulmonary disease. The statistics for NT-proBNP concentrations in the reference study group are shown in the following table.

Reference Study Group

All				
	<55 yrs	55 - 64 yrs	65 - 74 yrs	≥ 75 yrs
Mean	45.7	57.3	101.4	441.9
SD	50.2	59.2	57.0	904.0
Median	24.7	29.2	81.1	167.2
95 th Percentile	130.5	202.3	218.9	1707.0
% < 125 pg/mL	95%	87%	75%	-
% < 450 pg/mL	-	-	-	79%
N	163	15	8	122

Males				
	<55 yrs	55 - 64 yrs	65 - 74 yrs	≥ 75 yrs
Mean	39.6	70.7	116.4	518.0
SD	51.1	55.6	-	994.7

Median	15.0	32.0	116.4	162.1
95 th Percentile	163.5	157.4	116.4	1884.8
% < 125 pg/mL	95%	83%	100%	-
% < 450 pg/mL	-	-	-	73%
N	76	6	1	62

Females

	<55 yrs	55 - 64 yrs	65 - 74 yrs	≥ 75 yrs
Mean	51.0	48.3	99.2	363.3
SD	49.0	63.1	61.3	800.3
Median	36.6	16.7	81.1	167.2
95 th Percentile	130.5	202.3	218.9	1149.7
% < 125 pg/mL	94%	89%	71%	-
% < 450 pg/mL	-	-	-	85%
N	87	9	7	60

Disease Study Group

Blood samples were obtained from 234 patients diagnosed with CHF. The population included 70 women and 164 men. The descriptive statistics and New York Heart Association (NYHA) functional classes are provided below.

CHF Population – All

	<55 yrs	55 – 64 yrs	65 – 74 yrs	≥75 yrs
Mean	3872.0	3980.6	4894.9	5685.2
SD	6679.6	9543.7	8982.4	10090.8
Median	1416.7	938.2	1977.3	3040.1
95 th Percentile	19908.5	22270.3	13870.7	14196.0
% > 125 pg/mL	86%	83%	95%	-
% > 450 pg/mL	-	-	-	93%
N	56	53	58	67

CHF Population – Males

	<55 yrs	55 – 64 yrs	65 – 74 yrs	≥75 yrs
Mean	4050.3	4356.6	5076.5	6446.7
SD	6966.5	10754.3	10187.1	10806.6
Median	1660.9	1068.5	1732.7	3600.2
95 th Percentile	27839.2	22270.3	13827.9	14196.0
% > 125 pg/mL	87%	89%	93%	-
% > 450 pg/mL	-	-	-	92%
N	39	35	42	48

CHF Population – Females

	<55 yrs	55 – 64 yrs	65 – 74 yrs	≥75 yrs
Mean	3463.1	3249.5	4418.1	3761.5
SD	6152.5	6814.6	4754.6	7935.4
Median	714.1	530.6	2172.2	1098.9
95 th Percentile	19908.5	29255.0	13870.7	34234.1
% > 125 pg/mL	82%	72%	100%	-
% > 450 pg/mL	-	-	-	95%
N	17	18	16	19

CHF Population – All

NYHA Functional Class	All CHF	NYHA I	NYHA II	NYHA III	NYHA IV
Mean	4669.3	1674.8	2827.1	6057.0	10925.6
SD	8944.9	2355.7	3788.3	11432.4	13583.9
Median	1752.5	713.6	1201.7	2947.9	4823.8
5 th Percentile	84.9	54.9	34.9	171.8	91.8
95 th Percentile	16208.0	6407.7	9756.5	13827.9	33868.8
% > Cutoff	89%	80%	87%	99%	90%
Minimum	17.2	17.2	17.9	129.6	70.8
Maximum	74518.8	10400.1	22270.3	74518.8	64278.1
N	234	54	74	75	31

CHF Population – Males

NYHA Functional Class	All CHF	NYHA I	NYHA II	NYHA III	NYHA IV
Mean	5079.8	1869.8	3245.2	6792.7	13995.2
SD	9804.7	2508.4	3931.6	12699.9	16702.4
Median	2165.8	1068.5	2097.7	3444.7	6240.2
5 th Percentile	72.0	62.9	34.9	172.6	70.8
95 th Percentile	15602.8	6407.7	9756.5	13827.9	64278.1
% > Cutoff	90%	84%	89%	98%	88%
Minimum	17.2	17.2	29.1	159.9	70.8
Maximum	74518.8	10400.1	22270.3	74518.8	64278.1
N	164	43	52	53	16

CHF Population – **Females**

NYHA Functional Class	All CHF	NYHA I	NYHA II	NYHA III	NYHA IV
Mean	3707.4	912.2	1838.9	4284.6	7651.4
SD	6461.3	1477.1	3300.4	7518.6	8617.0
Median	1044.7	375.7	683.7	2172.2	3915.5
5 th Percentile	84.9	38.0	84.9	137.3	91.8
95 th Percentile	16208.0	5200.0	9515.2	12753.3	29255.0
% > Cutoff	87%	64%	82%	100%	93%
Minimum	17.9	38.0	17.9	129.6	91.8
Maximum	34234.1	5200.0	13665.4	34234.1	29255.0
N	70	11	22	22	15

These results show that there is a relationship between the severity of the clinical signs and symptoms of CHF and the median NT-proBNP concentration.

N. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.