510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY TEMPLATE

A. 510(k) Number:

k072437

B. Purpose for Submission:

Antibody changed from polyclonal anti-NT-proBNP antibodies to monoclonal anti-NT-proBNP antibodies

C. Measurand:

N-terminal pro-brain natriuretic peptide

D. Type of Test:

Quantitative

E. Applicant:

Roche Diagnostics, Inc.

F. Proprietary and Established Names:

Elecsys proBNP II Immunoassay

G. Regulatory Information:

1. Regulation section:

   21 CFR 862.1117, B-type natriuretic peptide

   21 CFR 862.1150, calibrator, secondary

   21 CFR 862.1660, multi-analyte controls, all kinds (assayed and unassayed)

2. Classification:

   Class II, Class II and Class I, respectively

3. Product code:

   NBC

   JIT

   JJY

4. Panel:

   Chemistry (75)

H. Intended Use:

1. Intended use(s):

   See Indications for Use below

2. Indication(s) for use:

   Immunoassay for the in vitro quantitative determination of N-terminal pro-Brain natriuretic peptide in human serum and plasma. Elecsys proBNP II assay is used 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 or congestive heart failure. The test may also serve as an aid in the assessment of increased risk of cardiovascular events and mortality in patients at risk for heart failure who have stable coronary artery disease. The electrochemiluminescence immunoassay "ECLIA" is intended for use on Elecsys and cobas e immunoassay analyzers.

   The Elecsys PreciControl Cardiac II is used for quality control of specified immunoassays on the Elecsys and cobas e immunoassay analyzers.

   The Elecsys proBNP II CalSet is used for calibrating the quantitative Elecsys proBNP II assay on the Elecsys and cobas e immunoassay analyzers.

3. Special conditions for use statement(s):

   Prescription use only

4. Special instrument requirements:

   Roche Diagnostics Elecsys and cobas e immunoassay analyzers

   The kit can be used on Elecsys analyzers including the MODULAR ANALYTICS E170 (Elecsys module) and cobas e 411 and cobas e 601 analyzers.

The cobas e analyzers include the cobas e 411 and cobas e 601 analyzers. The cobas e 411 analyzer is an updated version of the Elecsys 2010 analyzer. The modifications did not alter the measurement components of the analyzer or how the results are calculated. The two systems are analytically identical. The cobas e 601 analyzer is an updated version of the MODULAR ANALYTICS E170 analyzer. The cobas e 601 and MODULAR ANALYTICS E170 are analytically identical.

I. Device Description:

The Elecsys® proBNP II reagent kit contains the following three reagents:

M: Streptavidin-coated microparticles, 1 bottle, 6.5 mL: streptavidin-coated microparticles, 0.72mg/mL; preservative

R1: Anti-NT proBNP-Ab-biotin, 1 bottle, 9mL: Biotinylated monoclonal anti-NT-proBNP antibody (mouse) 1.1 µg/mL; phosphate buffer 40 mmol/L, pH 5.8; preservative

R2: Anti-NT-proBNP-Ab-Ru(bpy), 1 bottle, 9mL: monoclonal anti-NT-proBNP antibody (sheep) labeled with ruthenium complex 1.1 µg/mL; phosphate buffer 40 mmol/L, pH 5.8; preservative

J. Substantial Equivalence Information:

1. Predicate device name(s):

   Elecsys proBNP Immunoassay

2. Predicate K number(s):

   k051382

   Additional predicate k numbers for Elecsys proBNP Immunoassay are k022516 and k032646

3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Intended Use	Immunoassay for the in vitro quantitative determination of N-terminal pro-Brain natriuretic peptide in human serum and plasma. Elecsys proBNP II assay is used as an aid in the diagnosis of individuals suspected of having congestive heart failure. The test is further	same

Similarities		
Item	Device	Predicate
	indicated for the risk stratification of patients with acute coronary syndrome or congestive heart failure. The test may also serve as an aid in the assessment of increased risk of cardiovascular events and mortality in patients at risk for heart failure who have stable coronary artery disease.	
Detection protocol	Electrochemiluminescent	same
Sample type	Serum and plasma	same
Result interpretation	125 pg/mL for patients <75 years; 450 pg/mL for patients >75 years	same
Measuring Range	5-35,000 pg/mL	same

Differences		
Item	Device	Predicate
Antibody in R1 reagent	Biotinylated monoclonal anti-NT-proBNP mouse antibody	Biotinylated polyclonal anti-NT-proBNP sheep antibody
Antibody in R2 reagent	Ruthenylated monoclonal anti-NT-proBNP sheep antibody	Ruthenylated polyclonal anti-NT-proBNP sheep antibody
Calibrator	Elecsys proBNP II CalSet	Elecsys proBNP CalSet
Controls	Elecsys PreciControl Cardiac II	Elecsys PreciControl Cardiac

K. Standard/Guidance Document Referenced (if applicable):
- Class II Special Controls Guidance Document for B-Type Natriuretic Peptide Premarket Notifications: Final Guidance for Industry and FDA Reviewers (11/30/2000)
- CLSI EP6-A Evaluation of the Linearity of Quantitative Measurement
- CLSI - Evaluation of Precision Performance of Clinical Chemistry Devices - EP05-A2
- CLSI - Protocols for Determination of Limits of Detection and Limits of Quantitation - EP17-A

L. Test Principle:
The Elecsys® proBNP II Test System is an electrochemiluminescence immunoassay (ECLIA). The test uses the sandwich principle. In the first incubation, antigen in the sample, a biotinylated monoclonal NT-proBNP-specific antibody and a monoclonal NT-proBNP-specific antibody labeled with a ruthenium complex form a sandwich complex. In the second incubation, after addition of streptavidin labeled micro-particles, the complex produced is bound to the solid phase via biotin-streptavidin interaction. The reaction mixture is aspirated into the measuring cell where the

microparticles are magnetically captured onto the surface of the electrode. Unbound substances are then removed with ProCell. Application of a voltage to the electrode then induces chemiluminescent emission which is measured by a photomultiplier. Results are determined via a calibration curve. This curve is instrument-specifically generated by a 2-point calibration and a master curve provided via the reagent barcode.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

    a. Precision/Reproducibility:

    Precision was determined using Elecsys reagents, pooled human sera, and controls in a modified protocol (EP5-A) of the CLSI (Clinical and Laboratory Standards Institute; formerly NCCLS): 6 times daily for 10 days (n = 60); within-run precision on MODULAR ANALYTICS E170 analyzer, n = 21. The following results were obtained:

Elecsys 1010/2010 and **cobas e** 411 analyzers							
	Mean		Within-run precision			Total precision	
Sample	Mean		SD		CV	SD		CV
	pg/mL	pmol/L	pg/mL	pmol/L	%	pg/mL	pmol/L	%
HS[b] 1	44.0	5.19	1.84	0.22	4.2	2.02	0.24	4.6
HS 2	126	14.9	3.06	0.36	2.4	3.23	0.38	2.6
HS 3	2410	284	31.7	3.74	1.3	44.2	5.22	1.8
HS 4	33,606	3966	922	109	2.7	1288	152	3.8
PC[c] Cardiac II 1	82.0	9.68	2.11	0.25	2.58	2.27	0.27	2.8
PC Cardiac II 2	2318	274	27.3	3.22	1.18	36.6	4.32	1.6

b) HS = human serum
c) PC = PreciControl

MODULAR ANALYTICS E170 and **cobas e** 601 analyzers				
	Within-run precision			
Sample	Mean		SD		CV
	pg/mL	pmol/L	pg/mL	pmol/L	%
HS 1	64	7.55	1.21	0.14	1.9
HS 2	124	14.6	1.82	0.22	1.5
HS 3	14,142	1669	182	21.5	1.3
PC Cardiac II 1	77.0	9.09	1.41	0.17	1.8
PC Cardiac II 2	2105	248	24.8	2.92	1.2

MODULAR ANALYTICS E170 and **cobas e** 601 analyzers				
	Total precision			
Sample	Mean		SD		CV
	pg/mL	pmol/L	pg/mL	pmol/L	%
HS 1	46	5.43	1.44	0.17	3.1
HS 2	125	14.75	3.43	0.40	2.7
HS 3	32,805	3871	888	105	2.7
PC Cardiac II 1	77.0	9.09	2.12	0.25	2.7
PC Cardiac II 2	2170	256	59.4	7.01	2.7

An additional precision study was performed on the Elecsys 2010 on two samples with mean concentrations of 341.7 and 754.4 pg/mL which bracket the cutoff of 450 pg/mL for patients 75 years and older. Within run precision was determined for 21 replicates of each sample with CV 1.2% and 1.0% respectively. Total precision for the two samples was 2.3 % CV and 3.4 % respectively.

b. *Linearity/assay reportable range:*
The assay reportable range is from 5-35,000 pg/mL. In the first study, the linearity was evaluated by diluting four patient samples with varying amounts

of pro-BNP-free serum (10 concentrations per sample) and measured in triplicate on the MODULAR ANALYTICS E170 analyzer. The sponsor provided the data for 4 samples. Each sample was diluted with varying amounts of proBNP-free serum (11 concentrations per sample). Analysis of the linearity data was performed according to CLSI EP6-A. Because the precision data is proportional to concentration, the calculations were done on percentage difference scale rather than absolute difference scale. If the device is without measurement error and linear, then the percent recovery should be 1.00 (100%). For 44 measurements of recoveries, the assumption about constant standard deviation through entire range is acceptable. According to EP6-A, the linearity data should be analyzed with regard to linear, quadratic and cubic polynomials. The best fitted linear model for the percent recoveries was $0.9604 - 9E\text{-}07x$. The data demonstrated that the E170 analyzer was linear from 5 – 35,000 pg/mL with deviation from linearity not more than 15%.

In the second study, six patient samples were diluted with varying amounts of diluent (10 concentrations per sample) and measured in triplicate on the Elecsys 2010 analyzer. The samples ranged from 2.3 – 53,849 pg/mL. The data was analyzed according to CLSI EP6-A. Because the precision data is proportional to concentration, the calculations were done on percentage difference scale rather than absolute difference scale. If the device is without measurement error and linear, then the percent recovery should be 1.00 (100%). For 60 measurements of recoveries, the assumption about constant standard deviation through entire range is acceptable. According to EP6-A, the linearity data should be analyzed with regard to linear, quadratic and cubic polynomials. The best fitted linear model for the percent recoveries was $0.933 - 1E\text{-}07x$. The data demonstrated that the Elecsys 2010 analyzer was linear from 5 – 35,000 pg/mL with deviation from linearity not more than 10 %.

Hook effect studies demonstrated no effect up to 300,000 pg/mL.

c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*

Elecsys proBNP II CalSet

Summary of traceability: Elecsys proBNP II CalSet is traceable gravimetrically to an in house reference prepared by weighing pure synthetic NT-proBNP (1-76) into an equine serum matrix.

On board/open vial stability: Stability of the reconstituted Elecsys proBNP II CalSet for the Elecsys proBNP II assay was determined by studies on Elecsys 2010 analyzer. Onboard stability of Elecsys proBNP II CalSet after reconstitution is claimed for up to 5 hours at 20 – 25°C. Reconstituted calibrators were stored at 4 °C for two weeks and within this period they were

incubated at 32 °C for 5 hours. Each calibrator was tested in two-fold determinations. The signal percent recovery was within ± 10%.

Storage at -20°C: Elecsys proBNP II CalSet is claimed to be stable for three months at -20°C. Reconstituted calibrators were incubated for three months at -20°C and tested twice subsequently to determine the % recovery relative to the calibrators stored at 4°C. The signal percent recovery was within ± 10%.

Real-Time Stability: Elecsys proBNP II CalSet kits were stored at 2-8°C for 19 months. Real-time stability claims were established by testing the aged Calibrators with Elecsys PreciControl Cardiac II in two-fold determinations. Percent recovery was determined relative to the assigned target value. The shelf-life of Elecsys proBNP II CalSet is claimed to be 18 months with the kit unopened and stored at 2-8°C. The percent recovery was within ± 10%.

PreciControl Cardiac II:

On board /open vial stability: Onboard stability of Elecsys PreciControl Cardiac II after reconstitution was determined by studies on Elecsys 2010 analyzer. Onboard stability is claimed for up to 3 hours at 20 – 25°C. Reconstituted controls I and II were kept for three days at 4°C. Once per day they were stored at 22 °C for one hour. Each PreciControl was tested in two-fold determinations. The signal percent recovery was within ± 10%.

Stability after at -20°C storage: Elecsys PreciControl Cardiac II is claimed to be stable for 3 months at -20°C. Reconstituted controls I and II were stored for 3 months at -20°C and tested with two-fold determinations to determine the % recovery with reference to the calibrators stored at 4°C. The signal percent recovery was within ± 10%.

Real-Time Stability: Elecsys PreciControl Cardiac II kits were stored at 2-8°C and measured at determined time intervals for 19 months. Each PreciControl was tested in two-fold determinations and % recovery was determined to the assigned target value. The shelf-life of Elecsys PreciControl Cardiac II is claimed to be 18 month with the kit unopened and stored at 2-8°C. The percent recovery was within ± 10%.

Summary of value assignment: PreciControl Cardiac II values are assigned using a minimum of 3 Elecsys 1010 analyzers, 3 Elecsys 2010/cobas e 411 and 3 Elecsys MODULAR ANALYTICS E170 / cobas e 601. A minimum of 6 runs are performed on each analyzer platform. Two-fold determinations are made for each sample. The target value is then calculated as the median of the determined values. Target ranges for PreciControl Cardiac II are established at ± 12% of the target value. Lot specific target values are established.

d. Detection limit:

The limit of blank (LoB) was evaluated on the Elecsys 2010 and MODULAR ANALYTICS E170 analyzers according to CLSI EP17-A guidelines. Five human serum samples with low analyte concentration were measured on 2 analyzers in 3 runs per instrument. The samples were measured in duplicate in each run (n = 30). The LoD was determined to be 1.725 pg/mL for the Elecsys 2010 and 3.62 pg/mL for the E170. The labeling includes values for the limit of detection and limit of quantitation.

The limit of detection (LoD) was evaluated on the Elecsys 2010 and MODULAR ANALYTICS E170 analyzer according to CLSI EP 17-A guidelines. Five human serum samples with low analyte concentration were measured on analyzers in 3 runs per instrument. The samples were measured in duplicate in each run (n = 30). The limit of detection is determined based on the limit of blank and the standard deviation of low concentration samples. The LoD was determined to be 2.83 pg/mL for the Elecsys 2010 and 4.66 pg/mL for the E170. The sponsor claims a LoD of 5 pg/mL.

The limit of quantitation (LoQ) was determined on the Elecsys 2010 and E170 by measuring the concentration which corresponds to an interassay CV of 20 %. To determine the LoQ for the Elecsys 2010, 8 samples (6 human serum pools and 2 controls) with concentrations ranging from 2.39 to 2367.2 were tested once per day for 10 days. The mean standard deviation and CV for each sample was calculated. The concentration which would generate a 20% CV was determined to be < 14.0 pg/mL. For the E170, 10 samples (8 human serum pools and 2 controls) with concentrations ranging from 7.62 to 2168.8 were tested once per day for 10 days. The mean, standard deviation and CV for each sample were calculated. The concentration which would generate a 20 % CV was determined to be < 7.62 pg/mL. The sponsor claims the LoQ of 50 pg/mL.

e. Analytical specificity:
The assay is unaffected by icterus (bilirubin < 428 μmol/L or < 25 mg/dL), hemolysis (Hb < 0.621 mmol/L or < 1.0 g/dL), lipemia (intralipids < 17.1 mmol/L or < 1500 mg/dL), and biotin < 82 nmol/L or < 30 ng/mL.
In patients receiving therapy with high biotin doses (i.e. > 5 mg/day), no sample should be taken until at least 8 hours after the last biotin administration. No interference was observed from rheumatoid factors up to a concentration of 1500 IU/mL. In vitro tests were performed on 51 commonly used pharmaceuticals. No interference with the assay was found. In rare cases, interference due to extremely high titers of antibodies to streptavidin and ruthenium can occur.

f. *Assay cut-off:*

See expected values below

2. Comparison studies:

a. *Method comparison with predicate device:*

Accuracy was evaluated in a study comparing the values obtained with the Elecsys proBNP II assay to the values obtained with the Elecsys proBNP assay on the Elecsys 2010 analyzer. A total of 1551 samples were tested from three subgroups: Subgroup A (USA, 207 samples), Subgroup B (Europe, 328 samples) and Subgroup C (Europe, 1016 samples).

A comparison of the Elecsys proBNP II assay (y) with the Elecsys proBNP assay (x) using 1551 clinical samples with values ranging from 5 to 32,627 pg/mL gave the following correlations (pg/mL):

Passing/Bablok	Linear regression
y = 0.979x - 0.314	y = 1.00x - 21.9
τ = 0.932	r = 0.996

The sponsor provided concordance testing on reference and diseased group samples determined by comparing the results from the modified Elecsys proBNP II assay and the Elecsys proBNP assay. The concordance testing was performed on a subset of samples from the USA subgroup (Subgroup A), a subset of samples from Subgroup B and a subset of blood donor samples blood samples from Subgroup C. The reference group study examined samples from diabetic and hypertensive patients and from blood donors. The disease study group consisted of samples from each of the New York Heart Association (NYHA) classes (I – IV). The cut-offs of 125 pg/mL for patients < 75 years of age, and 450 pg/mL for patients ≥ years of age were used in the concordance determination. Concordance for the reference groups were 97.6 % for diabetics and hypertensives (94/96) and 99.0% for blood donors (607/613). Concordance for the disease group was 99% for Class I (142/143), 100% for Class II (159/159), 100 % for Class III (66/66), and 100% for Class IV (11/11).

In addition, concordance was evaluated in a study comparing the values obtained with the Elecsys proBNP II assay to the values obtained with the Elecsys proBNP assay using the samples from the USA Subgroup (Subgroup A). These samples were repository samples from the clinical studies performed in support of k022516, Elecsys proBNP assay. A comparison of

the Elecsys proBNP II assay (y) with the Elecsys proBNP assay (x) using the 207 clinical samples with values ranging from 5 to 11,258 pg/mL gave the following correlations (pg/mL):

Passing/Bablok   Linear regression
y = 0.983x + 1.914   y = 0.926x + 23.8
τ = 0.9708   r = 0.999

b. *Matrix comparison:*

The following sample types were tested on the Elecsys 2010 and cobas e 601 analyzers and found acceptable:
Serum collected using standard sampling tubes or tubes containing a separating gel, Li-, NH4-heparin and K2-, K3-EDTA plasma.
The acceptance criteria were: recovery within 90-110% of serum value or slope 0.9-1.1, intercept within <± 2 x analytical sensitivity (LDL) and coefficient of correlation > 0.95. The matrix comparison information using Passing/Bablock regression is presented in the table below:

	Serum vs. Li-heparin	Serum vs. NH4-heparin	Serum vs. K2-EDTA	Serum vs. K3-EDTA
Range (pg/mL)	11.5 – 18,675	56.8 – 19,207	15.56 – 27,448	57.77 – 19,825
N	30	22	58	22
Slope	1.000	0.9838	0.9986	0.994
y-intercept	-0.2236	0.2646	-4.0607	-5.288
R	1.000	0.9913	0.9722	0.9740
% recovery (mean)	100.4%	99.1%	100.9%	97.1%

3. Clinical studies:

a. *Clinical Sensitivity:*

Previously established for k022516 as shown in the table below:

Sensitivity and Specificity vs. Age and Gender

Males	< 45 years	45-54 years	55-64 years	65-74 years	75 + years	< 75 years
% Sensitivity	81.6	88.2	89.6	91.7	86.5	89.0
95% confidence interval	68.0-91.24	81.27-93.24	84.47-93.42	85.58-95.77	74.21-94.47	85.95-91.58
% specificity	95.7	93.3	87.8	86.7	88.9	90.0
95% confidence interval	78.05-99.89	89.07-96.31	82.33-91.99	75.59-92.07	77.37-95.81	87.14-92.32

Females	< 45 years	45-54 years	55-64 years	65-74 years	75 + years	< 75 years
% Sensitivity	86.7	90.5	89.3	94.3	81.8	90.6
95% confidence interval	59.54-98.34	69.62-98.83	78.12-95.97	80.84-99.30	64.54-93.02	84.08-95.02
% specificity	84.9	85.5	79.9	57.8	87.9	76.7
95% confidence interval	68.1-94.89	80.64-89.53	74.52-84.63	50.21-65.09	77.51-94.62	73.47-79.72

  b. *Clinical specificity:*

   See clinical sensitivity above.

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

4. Clinical cut-off**:**

   See expected values below.

5. Expected values/Reference range:

   Previously established in k022516 as: 125 pg/mL for patients younger than 75 years and 450 pg/mL for patients 75 years and older.

N. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

O. Conclusion:

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