510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K220456

B Applicant

Roche Diagnostics

C Proprietary and Established Names

Elecsys FT4 IV

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
CEC	Class II	21 CFR 862.1695 - Free Thyroxine Test System	CH - Clinical Chemistry

II Submission/Device Overview:

A Purpose for Submission:

New device

B Measurand:

Free Thyroxine (free T4)

C Type of Test:

Quantitative, electrochemiluminescent immunoassay (ECLIA)

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

Assay for the in vitro quantitative determination of free thyroxine in human serum and plasma. Measurements obtained by this device are used in the diagnosis and treatment of thyroid disease.

The electrochemiluminescence immunoassay "ECLIA" is intended for use on the cobas e immunoassay analyzers.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

For in vitro diagnostic use

D Special Instrument Requirements:

Cobas e 411 Immunoassay Analyzer

IV Device/System Characteristics:

A Device Description:

The Elecsys FT4 IV contains the following components:

M: Streptavidin-coated microparticles 0.72 mg/mL; preservative

R1: Anti-T4-antibody; Monoclonal anti T4 antibody (rabbit) labeled with ruthenium complex 75 ng/mL; phosphate buffer 100 mmol/L, pH 7.0; preservative

R2: T4-biotin; Biotinylated T4 2.5 ng/mL; phosphate buffer 100 mmol/L, pH 7.0; preservative

B Principle of Operation:

The assay is an electrochemiluminescence immunoassay with an assay time of 18 minutes. During the first incubation, 15 µL of sample is combined with a T4-specific antibody labeled with a ruthenium complex. During the second incubation, biotinylated T4 and streptavidin-coated microparticles are added, the still-free binding sites of the labeled antibody become occupied, with formation of an antibody-hapten complex. The entire complex becomes bound to the solid phase via interaction of biotin and streptavidin. 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 which is generated by 2-point calibration against the master curve for that reagent lot.

V Substantial Equivalence Information:

A Predicate Device Name(s):

Elecsys FT4 II Assay

B Predicate 510(k) Number(s):

K131244

C Comparison with Predicate(s):

Device & Predicate Device(s):	K220456	K131244
Device Trade Name	Elecsys FT4 IV	Elecsys FT4 II
General Device Characteristic Similarities		
Intended Use/Indications For Use	For the in vitro quantitative determination of free thyroxine in human serum and plasma. Measurements obtained by this device are used in the diagnosis and treatment of thyroid disease.	Same
Analyte	Free thyroxine	Same
Sample Type	Serum and plasma	Same
Methodology	Quantitative ECLIA	Same
General Device Characteristic Differences		
Antibodies used in R1	Ruthenylated monoclonal T4-specific rabbit antibody	Ruthenylated polyclonal T4-specific sheep antibody
Buffer composition R2	N-Biotinylsarcosine (Biotin-derivate)	D-Biotin
Biotin Tolerance	≤ 1200 ng/mL	≤ 20 ng/mL

VI Standards/Guidance Documents Referenced:

CLSI EP05-A3: Evaluation of Precision of Qualitative Measurement Procedures; Approved Guideline – Third Edition.
CLSI EP06 2nd Edition: Evaluation of the Linearity of Quantitative Measurement Procedures
CLSI EP07 3rd Edition: Interference Testing in Clinical Chemistry
CLSI EP09c 3rd Edition: Measurement Procedure Comparison and Bias Estimation Using Patient Samples
CLSI EP17-A2: Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures; Approved Guideline – Second Edition.
CLSI EP28-A3c: Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory; Approved Guideline – Third Edition
CLSI EP37 1st Edition: Supplemental Tables for Interference Testing in Clinical Chemistry

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

A precision study was performed based on CLSI EP05-A3 using one cobas e 411 analyzer. Six human serum samples and two levels of control material were assayed in duplicate, 2 runs per day for 21 days using a single reagent lot (n=84), results are summarized in the table below.

Sample	n	Mean (ng/dL)	Repeatability (Within-Run)		Intermediate Precision	
			SD (ng/dL)	CV (%)	SD (ng/dL)	CV (%)
Human serum 1	84	0.124	0.003	2.5	0.007	5.6
Human serum 2	84	0.515	0.006	1.1	0.012	2.3
Human serum 3	84	0.979	0.010	1.1	0.019	2.0
Human serum 4	84	1.82	0.017	1.0	0.031	1.7
Human serum 5	84	3.50	0.043	1.2	0.074	2.1
Human serum 6	84	6.85	0.117	1.7	0.181	2.6
PC Universal 1	84	1.17	0.010	0.9	0.021	1.7
PC Universal 2	84	3.09	0.037	1.2	0.064	2.1

The sponsor provided data to support that device results are similar between lots.

2. Linearity:

A linearity study was conducted according to CLSI EP06-2nd edition. Three linearity panels were prepared by spiking T4 into human serum samples to obtain an analyte concentration above the upper limit of the measuring interval. A depleted human serum sample with no free

T4 was used as the sample blank. The high and blank samples were mixed to prepare a dilution series with a total of 13 levels ranging from 0.08-9.64 ng/dL. Expected concentrations were assigned using the Elecsys FT4 II assay on the cobas e411 analyzer. Samples were tested by the candidate device using one cobas e 411 analyzer. The data was analyzed using first-order Weight Least Squares regression (WLS) without intercept. The results support the claimed measuring interval of 0.101 – 7.777 ng/dL.

3. Analytical Specificity/Interference:

Exogenous Interferents
Exogenous interference studies were performed according to the CLSI EP07 3rd Edition guideline. Two serum samples containing a low (1.5 ng/dL) and high (5 ng/dL) concentration of free T4 were assayed using the cobas e 411 analyzer. Results from the spiked samples were compared to results from the same serum sample pools without addition of the potential interferents (control samples). Significant interference was defined as more than ± 10% bias from the test results of the spiked sample when compared to the control sample. The following substances did not cause interference at the concentrations listed below:

Drug	**Highest concentration at which no significant interference was observed (µg/mL)**
Acetaminophen	156
Acetylcysteine	150
Acetylsalicylic Acid	30
Ampicillin	75
Ascorbic acid	52
Carbimazole	18
Cefoxitin	750
Cyclosporine	1.80
Doxycycline	18
Heparin	3300 IU/L
Ibuprofen	164
Itraconazole	15.0
Levodopa	7.50
Methyldopa	22.5
Metronidazole	123.0
Perchlorate	600
Phenylbutazone	80.3
Rifampicin	48.0
Theophylline	60
Thiamazole	80
Carbimazole	18
Levothyroxine-Na	0.25

Drug	Highest concentration at which no significant interference was observed (µg/mL)
Liothyronine	0.060
Thiamazole	80
Propylthiouracil	300
Perchlorate	600
Propranolol	120
Amiodarone	200
Prednisolone	100
Hydrocortisone	200
Octreotide	0.3
Furosemide	3.5
Liothyronine	0.02
Potassium Iodide	150
Lithium	540
Phenytoin	13.5
Carbamazepine	9

The following is included in the labeling:

- The drugs furosemide, carbamazepine, phenytoin, and levothyroxine sodium (L-T4, synthetic levothyroxine) caused elevated FT4 recovery at the daily therapeutic concentration.
- Any influence that might affect the binding behavior of the binding proteins can alter the result of the FT4 tests (e.g., drugs, NTIs (Non-Thyroid Illness)), patients suffering from FDH (Familial Dysalbuminemic Hyperthyroxinemia) or increased TBG in pregnancy.
- The test cannot be used in patients receiving treatment with lipid-lowering agents containing D-T4. If the thyroid function is to be checked in such patients, the therapy should first be discontinued for 4-6 weeks to allow the physiological state to become re-established.
- In rare cases, interference due to extremely high titers of antibodies to analyte-specific antibodies, streptavidin or ruthenium can occur.

Cross-Reactivity

The candidate device was tested for potential cross-reactivity. Human serum samples were used to prepare samples with free T4 concentrations of approximately 1.65 ng/dL, 2.66 ng/dL, 5.2 ng/dL and 8.3 ng/dL that were then spiked with the cross-reactants listed below at the concentrations listed below. Samples were assayed in duplicate on the cobas e 411 analyzer. For all substances tested, no significant cross-reactivity was defined as recovery ± 10% of initial value. The following cross-reactants demonstrated no reactivity at the concentrations tested:

Cross-reactant	Cross-reactant concentration tested (ng/dL)
L-T3	50,000
D-T3	50,000
rT3	190,000
3-iodo-L-tyrosine	10,000,000
3,5-diiodo-L-tyrosine	10,000,000
3,3',5-triiodothyroacetic acid	100,000
3,3',5,5'-tetraiodothyroacetic acid	100,000

Biotin interference
Biotin interference was evaluated by spiking serum samples with low, medium, and high concentrations of free T4 with 0, 75, 150, 300, 500, 750, 1050, 1200, 2400, 3600 and 10800 ng/mL biotin. Results from the spiked samples were compared to results from the same serum sample pools without addition of the potential interferents (control samples). Significant interference was defined as more than ± 10% bias from the test results of the spiked sample when compared to the control sample. The maximum value with no interference observed was 3600 ng/mL. The claim of the package insert is no interference up to 1200 ng/mL.

The labeling includes the following:
Specimens that contain biotin at a concentration of 1200 ng/mL demonstrate less than or equal to 10 % bias. Pharmacokinetic studies have shown that serum concentrations of biotin can reach up to 355 ng/mL within the first hour after biotin ingestion for subjects consuming supplements of 20 mg biotin per day and up to 1160 ng/mL for subjects after a single dose of 300 mg biotin.

4. Assay Reportable Range:

   See Linearity section above.

5. Traceability, Stability, Expected Values (Controls, Calibrators, or Methods):

   The Elecsys FT4 IV assay is traceable to the Roche Elecsys FT4 II assay (K131244). The Elecsys FT4 II assay is traceable to the Enzyme Test FT4 which has been standardized using equilibrium dialysis.

6. Detection Limit:

   Limit of Blank (LoB), Limit of Detection (LoD), and Limit of Quantitation (LoQ) were evaluated based on the CLSI guideline EP17-A2.

   To calculate LoB, one cobas e 411 analyzer was used with three reagent lots to assay free T4-depleted human serum in six runs with ten replicates per run over three days (n=60

measurements/lot). LoB was determined to be 0.02 ng/dL per EP17-A2 as the linear interpolation of the 57th and 58th ranked observation.

To calculate LoD, one cobas e 411 analyzer was used with three reagent lots to assay five low-level free T4 serum samples. Each sample was measured in two replicates per run for 6 runs (one run per day, over multiple days). LoD was determined to be 0.04 ng/dL.

LoQ was determined by measuring ten low serum samples (diluted single donor sera with free T4-depleted serum), in five replicates per run, one run per day, over 5 days, using 3 lots of reagents (25 replicates per sample per reagent lot). LoQ is defined as the lowest analyte concentration that can be reproducibly measured with an intermediate precision CV of ≤ 20 % and was determined to be 0.101 ng/dL.

These studies support the claimed analytical measuring interval of 0.101 – 7.7 ng/dL.

7. Assay Cut-Off:

Not applicable

B Comparison Studies:

1. Method Comparison with Predicate Device:

A method comparison study was conducted by comparing the Elecsys FT4 IV assay on the cobas e 411 analyzer to the comparator, the Elecsys FT4 II on the cobas e 411 analyzer using a protocol based on CLSI EP09c-Ed3. A total of 121 serum samples were assayed using one lot of candidate and comparator reagents. Passing-Bablok and linear regression analysis results between the candidate device (dependent variable, y) and the predicate device (x, comparator), are shown below:

Linear regression	y = 1.039x - 0.034	r = 0.999
Passing-Bablok	y = 1.034x - 0.025	$_T$ = 0.967

2. Matrix Comparison:

Anticoagulants:
A matrix comparison study was conducted to compare lithium heparin plasma, K2-EDTA plasma, K3-EDTA plasma, and serum samples. 56 matched native samples (free T4 concentrations ranging from 0.11 – 7.27 ng/dL) were run in singleton on one cobas e 411 analyzer using one reagent lot. Results analyzed by Passing-Bablok regression are shown below:

Sample Type Comparison	Slope	Intercept (ng/dL)	Correlation Coefficient
Serum/Li-Heparin	1.008	-0.011	0.997
Serum/K2-EDTA	1.020	-0.017	0.998
Serum /K3 EDTA	1.018	-0.022	0.997

The results support that K2-EDTA, K3-EDTA, and lithium heparin plasma samples are suitable for use with the Elecsys FT4 IV assay.

C Clinical Studies:

1. Clinical Sensitivity:

   Not applicable

2. Clinical Specificity:

   Not applicable

3. Other Clinical Supportive Data (When 1. and 2. Are Not Applicable):

   Not applicable

D Clinical Cut-Off:

Not applicable

E Expected Values/Reference Range:

Euthyroid: 0.92-1.68 ng/dL

These values correspond to the 2.5th and 97.5th percentile of results from a total of 150 healthy test subjects studied.

VIII Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device.

IX Conclusion:

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