510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K221197

B Applicant

Ortho-Clinical Diagnostics

C Proprietary and Established Names

VITROS Immunodiagnostic Products Intact PTH II Reagent Pack

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
CEW	Class II	21 CFR 862.1545 - Parathyroid Hormone Test System	CH - Clinical Chemistry

II Submission/Device Overview:

A Purpose for Submission:

New device

B Measurand:

Intact Parathyroid Hormone (iPTH)

C Type of Test:

Quantitative Sandwich Immunoassay

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

VITROS Immunodiagnostic Products Intact PTH II Reagent Pack quantitatively measures intact parathyroid hormone (iPTH) in human serum and plasma (K2-EDTA, lithium heparin or sodium heparin) using the automated VITROS 3600 Immunodiagnostic System.

Intact PTH is indicated to aid in the differential diagnosis of hyperparathyroidism, hypoparathyroidism, differential diagnosis of hypocalcemia or hypercalcemia and for intraoperative measurement of iPTH levels.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

D Special Instrument Requirements:

VITROS 3600 Immunodiagnostic System

IV Device/System Characteristics:

A Device Description:

The device consists of the components described below.

Each reagent pack contains:

- 100 coated wells (mouse monoclonal anti-PTH antibody, 2 ug/mL)
- 7.4 mL assay reagent as buffer with bovine gamma globulin, bovine serum albumin, and antimicrobial agent
- 7.4 mL conjugate reagent (HRP-mouse monoclonal anti-PTH, 6 ug/mL) in buffer with bovine serum albumin and antimicrobial agent

<u>Materials Required but Not Provided</u>

The VITROS Intact PTH II Assay is designed for use in conjunction with the following VITROS Products:

- VITROS 3600 Immunodiagnostic System
- VITROS iPTH II Calibrators
- VITROS iPTH II Range Verifiers
- VITROS Immunodiagnostic Products Signal Reagent
- VITROS Immunodiagnostic Products Universal Wash Reagent
- VITROS Immunodiagnostic Products PTH Controls

B Principle of Operation:

The candidate device is a single step immunometric test for the quantitative measurement of intact parathyroid hormone (iPTH) in human serum and plasma. The test sample (80 µL) is first added to the Biotin-BSA, Streptavidin, and a biotin/anti-PTH antibody conjugate (mouse monoclonal) coated well, followed by assay reagent (sample diluent). Next, horseradish peroxidase (HRP)-labelled anti-PTH antibody conjugate (mouse monoclonal) is added to the well, followed by incubation. Unbound (HRP)-labelled anti-PTH antibody conjugate is then removed by washing. The bound HRP conjugate is measured by luminescent reaction. The HRP in the bound conjugate catalyzes the oxidation of the luminol derivative substrate, producing light. The amount of HRP conjugate bound is directly proportional to the concentration of iPTH present in the sample. The reaction is complete in approximately 18 minutes.

V Substantial Equivalence Information:

A Predicate Device Name(s):
Elecsys PTH Immunoassay and Elecsys PTH STAT Immunoassay

B Predicate 510(k) Number(s):

K070709

C Comparison with Predicate(s):

Device & Predicate Device(s):	K221197	K070709
Device Trade Name	VITROS Immunodiagnostic Products Intact PTH II Reagent Pack	Elecsys PTH Immunoassay and Elecsys PTH STAT Immunoassay
General Device Characteristic Similarities		
Intended Use	Quantitative measurement of iPTH	Same
Test Principle	Chemiluminescent immunoassay	Same
Sample Type	Human serum and plasma	Same
General Device Characteristic Differences		
Measuring Range	6.8-5000 pg/mL	1.20-5000 pg/mL

VI Standards/Guidance Documents Referenced:

- CLSI EP05-A3 Evaluation of Precision of Quantitative Measurement Procedures; Approved Guideline - Third Edition

- CLSI EP09c 3rd Edition Measurement Procedure Comparison and Bias Estimation Using Patient Samples

- CLSI EP37 1st Edition Supplemental Tables for Interference Testing in Clinical Chemistry

- CLSI EP17-A2 Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures; Approved Guideline - Second Edition

- CLSI EP06-2nd Edition Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline

- CLSI EP07 3rd Edition Interference Testing in Clinical Chemistry.

- ISO 15711- 2nd Edition In vitro diagnostic medical devices - Requirements for establishing metrological traceability of values assigned to calibrators, trueness control materials and human samples

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

A precision study was performed in accordance with CLSI EP05-A3. Pooled serum patient samples (spiked at higher concentrations) were tested using three reagent lots on two Vitros 3600 Immunodiagnostic Systems. Samples were assayed in duplicate using 2 runs per day over 20 days for a total of 80 replicates per sample per lot. Within lab precision was determined using a single reagent lot. Within-run (repeatability) and within-lab (total) precision results for a representative reagent lot on a single instrument are summarized below:

Mean (pg/mL)	Repeatability		Within Lab	
	SD (pg/mL)	CV%	SD (pg/mL)	CV%
16	0.2	1.1	0.4	2.7
83	0.9	1.1	1.5	1.8
157	1.8	1.1	3.2	2.0
751	9.3	1.2	13.6	1.8
2049	30.9	1.5	47.2	2.3
4786	74.7	1.6	113.9	2.4

2. Linearity:

A linearity study was performed in accordance with CLSI EP06 2nd Edition. A high and a low human serum pool were prepared by spiking stripped, filtered human serum. Thirteen (13) dilutions were created by mixing the high human serum pool with the low human serum pool. The samples spanned from 2.9 pg/mL to 5,200 pg/mL PTH. Five (5) replicates were

measured for each panel member using a single reagent lot on one VITROS 3600 Immunodiagnostic System. The data were analyzed using a weighted linear regression model. The deviation from linearity did not exceed 5.9%.

The results support the claimed measuring interval of 6.8 – 5000 pg/mL.

3. Analytical Specificity/Interference:

Interfering Substances:
An endogenous and exogenous interference study was performed in accordance with CLSI EP-07 3rd edition. Endogenous serum samples with low (~30 pg/mL) or high (~100 pg/mL) iPTH concentrations were spiked with potentially interfering substances. Percent interference was calculated using the following equation:

$$100 \times \frac{\text{(mean result of the test substance)} - \text{(mean result of the control sample)}}{\text{mean result of the control sample}}$$

Interference was defined by the sponsor as ≥ 10% difference from the control sample.

The interference study results are summarized below:

Endogenous Substance	Highest concentration tested at which no significant interference was observed
Bilirubin, conjugated	40 mg/dL
Bilirubin, unconjugated	40 mg/dL
Cholesterol	400 mg/dL
Fibrinogen	1000 mg/dL
Hemoglobin	1000 mg/dL
Human Anti-Mouse Antibodies (HAMA)	800 ug/L
Triglycerides, total	1500 mg/dL

Exogenous Substance	Highest concentration tested at which no significant interference was observed
Acetaminophen	15.6 mg/dL
Acetylcysteine	15.0 mg/dL
Albuterol (salbutamol)	0.0045 mg/dL
Alendronate sodium	33.21 µg/dL
Aliskiren	255 µg/dL
Alprazolam	0.0258 mg/dL
Amitriptyline HCl	0.0542 mg/dL

Exogenous Substance	Highest concentration tested at which no significant interference was observed
Amlodipine besylate	0.104 mg/dL
Amoxicillin	5.40 mg/dL
Ascorbic acid	5.25 mg/dL
Atorvastatin calcium trihydrate	0.162 mg/dL
Benazepril HCl	0.044 mg/dL
Biotin	3510 ng/mL
Caffeine	10.8 mg/dL
Calcitriol	0.432 µg/dL
Carbamazepine	4.5 mg/dL
Ceftriaxone disodium hemi (heptahydrate)	0.99 mg/dL
Cephalexin sodium	13.4 mg/dL
Cinacalcet Hydrochloride	0.0259 mg/dL
Ciprofloxacin	1.2 mg/dL
Clarithromycin	0.720 mg/dL
Cotinine	0.240 mg/dL
Dextran	2400 mg/dL
Dextromethorphan	0.00156 mg/dL
Digoxin	0.0039 mg/dL
Diphenhydramine HCl	0.0884 mg/dL
Dipyrone (4-methylaminoantipyrine)	3.30 mg/dL
Enalaprilat	0.0819 mg/dL
Epoetin alfa	20000 mU/mL
Equilin	1.50 mg/dL
Estrone	0.0297 µg/dL
Ethanol	600 mg/dL
Fluoxetine	0.142 mg/dL
Fosrenol (Lanthanum Carbonate)	0.300 µg/dL
Furosemide	1.59 mg/dL
Glyburide	0.072 mg/dL
Guaifenesin	0.450 mg/dL
Hydrochlorothiazide	0.113 mg/dL

Exogenous Substance	Highest concentration tested at which no significant interference was observed
Ibuprofen	21.9 mg/dL
L-dopa (Levodopa)	0.75 mg/dL
Levothyroxine	0.0429 mg/dL
Loratadine	0.0087 mg/dL
Naproxen sodium	39.3 mg/dL
Nifedipine	0.0588 mg/dL
Omeprazole	0.84 mg/dL
Oxycodone HCl	0.0362 mg/dL
Paricalcitol	0.750 µg/dL
Phenytoin	6.00 mg/dL
Prednisone	0.0099 mg/dL
Propranolol HCl	0.115 mg/dL
Pseudoephedrine	0.330 mg/dL
Ranitidine HCl	1.17 mg/dL
Rifampicin (Rifampin)	4.80 mg/dL
Salicylic Acid	2.86 mg/dL
Sodium Azide	100 mg/dL
Spironolactone	0.0555 mg/dL
Terazosin	0.0273 mg/dL
Triamterene	0.0585 mg/dL
Vancomycin hydrochloride	12.3 mg/dL
Warfarin sodium	8.03 mg/dL

Interference was observed for three (3) substances (cefoxitin sodium, rheumatoid factor and total protein) with the VITROS Immunodiagnostic Product Intact PTH II Reagent Pack. As shown below, bias ≥ 10% was reported for each substance, except rheumatoid factor at 100 pg/mL iPTH.

Interferent	iPTH Conc. (pg/mL)	Interferent Concentration	% Bias
Cefoxitin Sodium*	30	174.25 mg/dL	-11.4
Rheumatoid Factor		675 IU/mL	13.5
Total protein		11.8 g/dL	-14.3
Cefoxitin Sodium	100	174.25 mg/dL	-12.2
Rheumatoid Factor		900 IU/mL	6.3
Total protein		11.9 g/dL	-14.0

* The level of interference for Cefoxitin Sodium is within the therapeutic range for this compound and could lead to lower reported PTH concentrations for patients on this compound.

The following limitations are included in the labeling:

Heterophile as well as human anti-animal antibodies (most common human anti-mouse antibodies or HAMA) in serum or plasma of certain individuals are known to cause interference with immunoassays. The anti-animal antibodies may be present in blood samples from individuals regularly exposed to animals or who have received preparations of mouse monoclonal antibodies for diagnosis or therapy. Results inconsistent with clinical observations indicate the need for additional testing.

Patients taking Cefoxitin Sodium could have reported PTH concentrations that are negatively biased at levels indicated in the Known Interferences section.

Rheumatoid factor concentrations less than 450 IU/mL have demonstrated no observed interference. Rheumatoid factor at concentrations of 675 IU/mL and above have been shown to falsely elevate PTH test results.

Total protein at concentrations less than 9.4 g/dL have demonstrated no observed interference. Total protein at concentrations of 11.8 g/dL and above have been shown to falsely decrease PTH test results.

Cross-Reactivity:
Cross-reactivity studies were performed according to CLSI EP-07 3rd edition. Cross-reactivity was determined by spiking each cross-reactant into serum containing either no PTH (stripped serum) or 30 pg/mL endogenous PTH. Each test and control sample was measured in five (5) replicates using three reagent lots on one (1) instrument. Percent cross-reactivity was calculated using the following equation:

$$100 \times \frac{\text{(Mean result of spiked sample – Mean result of control sample)}}{\text{Spike concentration of cross-reactant}}$$

The cross-reactivity study results are summarized in the following table:

Test Substance	Concentration	Mean Cross-reactant Sample Result		% Cross-reactivity
		pg/mL	pmol/L	
Alkaline Phosphatase	120 ng/mL	*ND	*ND	*ND
β-Cross laps	10 ng/mL	*ND	*ND	*ND
Calcitonin	100,000 pg/mL	*ND	*ND	*ND
Osteocalcin	50 ng/mL	*ND	*ND	*ND
PTH 1-34	100,000 pg/mL	*ND	*ND	*ND
PTH 39-84	100,000 pg/mL	*ND	*ND	*ND
PTH 7-84	1000 pg/mL	613.2	65.0	61.3

***Not Detectable (ND). Concentration was below the measuring interval of the test (68– 5000 pg/mL).**

The following limitation is included in the labeling:

The VITROS Intact PTH II assay will detect non-intact PTH molecules, such as the large C-terminal PTH fragment 7-84. PTH fragments, including 7-84, may cause falsely elevated PTH results in patients with abnormal renal function as these patients may have various concentrations of PTH fragments in their blood. In patients with atypical renal function, interpret the PTH result with caution, and do not make patient management decisions on the PTH result alone. A study describing PTH fragmentation is provided in Lopez et al "Selected Reaction Monitoring Mass Spectrometric Immunoassay Responsive to Parathyroid Hormone and Related Variants."

High dose hook effect:

No high dose hook effect was observed for samples containing up to 837,800 pg/mL (88,807 pmol/L) iPTH.

4. Assay Reportable Range:

   See Linearity section above.

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

   The VITROS Immunodiagnostic Products Intact PTH II Reagent Pack is traceable to internal standards prepared with synthetic human iPTH.

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.

LoB was determined using five (5) samples containing no measurable PTH. Each of the 5 samples were measured 20 times over 5 days using 1 instrument and 3 reagent lots for a total of 100 replicates per reagent lot. The LoB was calculated non-parametrically by using the 95th percentile of 100 ranked observations for each lot.

LoD was determined using a non-parametric approach and included 5 low measurand serum sample pools. Each sample was measured in 50 replicates over 5 days using 1 instrument and 3 reagent lots for a total of 250 replicates per reagent lot. The LoD value was determined as the highest resultant value of the three lots.

LoQ was determined using the precision profile approach and included 5 low measurand serum sample pools with analyte concentrations ranging from 1.2 to 2.4 pg/mL. Each sample was measured in 50 replicates over 5 days using 1 instrument and 3 reagent lots for a total of 250 replicates per reagent lot. LoQ is defined as the lowest analyte concentration that can be measured with an intermediate precision CV of ≤ 20 %.

The LoB, LoD, and LoQ results are summarized below:

LoB	LoD	LoQ
0.8 pg/mL	1.2 pg/mL	1.2 pg/mL

7. Assay Cut-Off:

Not applicable.

B Comparison Studies:

1. Method Comparison with Predicate Device:

A method comparison study comparing the VITROS Immunodiagnostic Products Intact iPTH II Reagent Pack assay and the Roche Elecsys PTH assay (K070709) was conducted in accordance with CLSI EP09. A total of 206 serum samples were tested in singlicate over multiple days. Nineteen (19) of the 206 (9%) serum samples used were contrived. Passing Bablok regression analysis was performed, and the results are summarized below.

N	Sample range tested (pg/ml)	Slope	Intercept (pg/mL)	Correlation Coefficient (r)
206	7.7 - 4384	1.01	0.3	0.991

2. Matrix Comparison:

A matrix comparison study was conducted based on CLSI EP35. A total of 85 paired Serum, K2-EDTA plasma, lithium-heparin plasma (Li-Hep), and sodium heparin plasma (Na-Hep) samples were tested in duplicate each day of testing on two VITROS 3600 Immunodiagnostic Systems using 1 reagent lot per paired sample. A total of 3 reagent lots were used in the study. Passing Bablok regression analysis was performed. For each specimen, the first replicate result in the candidate matrix types (K2-EDTA plasma, Li-Hep and Na-Hep) was plotted against the mean result in the primary matrix type (serum). The results are summarized in the table below:

Comparison	Regression Equation	R
Serum vs. Li-Hep Plasma	y = 1.011x +0.6659	0.996
Serum vs. NaHep Plasma	y = 1.009x + 0.0441	0.994
Serum vs. EDTA Plasma	y = 0.988x + 0.1949	0.973

The study results support the sponsor's claim that human serum and plasma (EDTA, lithium heparin, sodium heparin) are acceptable sample types to be used with this 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):

Intra-Operative Study:

Intra-operative samples were collected and tested to establish the intra-operative claims of the candidate test. Serum from 32 parathyroidectomy subjects with primary hyperparathyroidism were assessed in singlicate using the VITROS Intact PTH II assay versus a comparator PTH assay FDA cleared for intra-operative use. The surgeons determined successful surgery (a surgery where a 50% or greater drop in intact PTH level from the pre-incision or pre-excision baseline values to the post-excision test result after the last parathyroid gland excision was observed based on the PTH assay used intraoperatively). Concordance between the comparator device used intraoperatively and the VITROS Immunodiagnostic Products Intact PTH II Reagent Pack assay is presented in the table below.

		Comparator Assay	
		Successful	Unsuccessful
VITROS Immunodiagnostic Products Intact PTH II Reagent Pack	Successful	29	0
	Unsuccessful	0	3

Primary Endpoint Positive Agreement = 29/29 = 100%
Primary Endpoint Negative Agreement = 3/3 = 100%
Primary Endpoint Overall Agreement = 32/32 = 100%

D Clinical Cut-Off:

Not applicable.

E Expected Values/Reference Range:

The reference range was assessed in accordance with CLSI EP28 using 134 serum samples from apparently healthy donors with normal calcium, magnesium, alkaline phosphatase, phosphate and TSH levels. Samples were analyzed in singlicate with one VITROS 3600 Immunodiagnostic System and four reagent lots. The results of the reference range study are shown below.

N	95% Reference Interval	
	Conventional Units (pg/mL)	Alternate Units (pmol/L)
134	14.5 - 79.4	1.5 - 8.4

F Other Supportive Instrument Performance Characteristics Data:

The sponsor provided data to support that spiked samples used in validation studies are comparable to native patient samples.

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.