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

A. 510(k) Number:
K040813

B. Purpose for Submission:
Labeling changes, Indications for Use modified

C. Analyte:
Parathyroid hormone,

D. Type of Test:
Quantitative

E. Applicant:
Nichols Institute Diagnostics

F. Proprietary and Established Names:
Nichols Advantage® Chemiluminescence Intact Parathyroid Hormone

G. Regulatory Information:

1. Regulation section:
   862.1545, Parathyroid hormone test system
2. Classification:
   Class II
3. Product Code:
   CEW
4. Panel:
   75

H. Intended Use:

1. Intended use(s):
   The Nichols Advantage® Chemiluminescence Intact Parathyroid Hormone Immunoassay is intended for use with the Nichols Advantage® Specialty System for the quantitative determination of intact parathyroid hormone in human EDTA plasma, heparinized plasma and serum. Measurements of parathyroid hormone levels are used in the differential diagnosis of hypercalcemia (abnormally high levels of calcium in the blood) and hypocalcemia (abnormally low levels of calcium in the blood) resulting from disorders of calcium metabolism. Measurements of intact parathyroid hormone levels are also used as an aid in monitoring therapeutic intervention of secondary hyperparathyroidism that frequently occurs in chronic kidney

disease. Assay results should be used in conjunction with other clinical data to assist the clinician in making individual patient management decisions.

2. Indication(s) for use:
   See Intended Use above
3. Special condition for use statement(s):

4. Special instrument Requirements:
   Nichols Advantage® Specialty System

I. Device Description:

One (1) cartridge contains the following reagents sufficient for 100 tests:

1. Streptavidin Coated Magnetic Particles
   One vial (2.8 mL) containing streptavidin coated magnetic particles in a buffered protein solution with ≤0.095% sodium azide and Proclin® 300
2. Acridinium Labeled Antibody Solution
   One vial (3.0 mL) containing acridinium ester-labeled goat polyclonal antibody to human intact PTH in a buffered protein solution with ≤0.095% sodium azide
3. Biotinylated Antibody Solution
   One vial (3.0 mL) containing biotinylated goat polyclonal antibody to human intact PTH in a buffered protein solution with ≤0.095% sodium azide and Proclin® 300
4. Assay Buffer
   One vial (5.6 mL) containing goat serum based solution with ≤0.095% sodium azide as preservative
5. One lot specific Intact PTH Master Curve Bar Code Card

J. Substantial Equivalence Information:

1. Predicate device name(s):
   Nichols Advantage® Chemiluminescence Intact Parathyroid Hormone
2. Predicate K number(s):
   K962598
3. Comparison with predicate:

Similarities		
Item	**Device**	**Predicate**
Test principle	Two-site chemiluminescence immunoassay	Same
Antibody	Two goat polyclonal antibodies to human intact PTH	Same
Separation system	Streptavidin coated magnetic particles	Same

Differences		
Item	**Device**	**Predicate**
Indications for Use	The Nichols Advantage® Chemiluminescence Intact Parathyroid Hormone Immunoassay is intended for use with the Nichols Advantage® Specialty System for the quantitative determination of intact parathyroid hormone in human EDTA plasma, heparinized plasma and serum. Measurements of parathyroid hormone levels are used in the differential diagnosis of hypercalcemia (abnormally high levels of calcium in the blood) and hypocalcemia (abnormally low levels of calcium in the blood) resulting from disorders of calcium metabolism. Measurements of intact parathyroid hormone levels are also used as an aid in monitoring therapeutic intervention of secondary hyperparathyroidism that frequently occurs in chronic kidney disease. Assay results should be used in conjunction with other clinical data to assist the clinician in making individual patient management decisions.	The Nichols Advantage® Chemiluminescence Intact Parathyroid Hormone Immunoassay assay is for use only with the Nichols Advantage® Specialty System for the quantitative measurement of intact parathyroid hormone concentrations in human serum and EDTA plasma.
Sample types	Serum, EDTA plasma, heparin plasma	Serum, EDTA plasma
Sample collection and storage instructions	EDTA and heparinized plasma may be stored refrigerated (2-8ºC) or room temperature (15-25º C) up to 48 hours. EDTA whole blood may be stored refrigerated (2-8 ºC) up to 48 hours before centrifugation. Serum should be rapidly processed and frozen within 2 hours of collection.	Centrifuge samples and freeze serum and plasma immediately.

K. Standard/Guidance Document Referenced (if applicable):
None referenced

L. Test Principle:
The Nichols Advantage Intact PTH assay is a two-site chemiluminescence immunoassay. Two goat polyclonal antibodies to human intact PTH are used. One antibody is coupled to biotin while the second antibody is labeled with acridinium ester for detection. Intact PTH is "sandwiched" between these antibodies. After an initial incubation period with the acridium labeled antibody, streptavidin coated magnetic particles and the capture antibody are added to the reaction mixture and a second incubation follows. Unbound labeled antibody is separated from the labeled antibody bound to the magnetic particles by aspiration of the reaction mixture and subsequent washing. The wells containing the washed magnetic particles are transported into the system luminometer, which automatically injects Trigger 1 and Trigger 2, initiating the chemiluminescence reaction. The light is quantitated by the luminometer and expressed as RLU. The amount of bound-labeled antibody is directly proportional to the concentration of intact PTH in the sample.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:
The precision (intra-assay variation) of the Nichols Advantage Intact PTH Assay was calculated from 20 replicate determinations on six levels of intact PTH in a single run using six human serum pools. The reproducibility (inter-assay) was calculated on three levels of intact PTH over 20 runs using three human serum pools. The % Coefficient of Variation is expressed as 90% Confidence level for the Variance.

Intra-Assay Variation

Mean PTH Value (pg/mL)	% Coefficient Variation	N
2.0	12.5	20
8.0	6.7	20
17.1	4.1	20
142	4.4	20
338	5.7	20
786	3.8	20

Inter-Assay Variation

Mean PTH Value (pg/mL)	% Coefficient Variation	N
33.7	9.2	20
86.7	7.0	20
229	7.5	20

b. Linearity/assay reportable range:
Previously established for K962598. The highest reportable value without dilution is the value of the highest point on the Master Curve (1800 pg/mL). Samples reading above the Master Curve should be diluted and repeated, or reported as greater than the highest value on the Master Curve. Samples with varying concentrations of intact PTH were diluted with Sample Diluent before placing onto the system, or diluted on-board the system. The results demonstrate linearity across the range of the assay.

c. Traceability (controls, calibrators, or method):
The PTH standards are prepared analytically on a mass basis from purified synthetic human intact PTH.

d. Detection limit:
Previously established for K962598. The limit of detection is estimated to be 1.0 pg/mL.

e. Analytical specificity:
Specificity and Cross-Reactivity
The antibody pair in the Nichols Advantage Intact PTH kit comprises a biotinylated antibody specific to epitopes within PTH 39-84, and a labeled acridinium ester antibody specific to epitopes on amino-terminal PTH 1-34. A sandwich-complex is formed when an intact PTH molecule or when an amino-terminal truncated PTH molecule bridges both antibodies. The following substances were added to the Intact PTH zero standard.

Cross-Reactant	Concentration of Cross-Reactant (pg/mL)	Observed value in zero standard (pg/mL)
PTH 1-34	300	N.D.
PTH 39-68	100,000	N.D.
PTH 53-84	100,000	N.D.
PTH 44-68	100,000	N.D.
PTH 39-84	100,000	1.3
PTH 7-84	1,250	1,127
PTHrP 1-40	80	N.D.

N.D. Not Detectable

f. Assay cut-off:
NA

2. Comparison studies:

a. Method comparison with predicate device:
The Nichols Advantage Intact PTH Assay (y) was compared to the Nichols Institute Diagnostics Intact PTH Immunoradiometric Assay (x). A population of 190 samples was assayed by each method. The range of values obtained using the Nichols Institute Diagnostics

Intact PTH Immunoradiometric Assay (x) were from 0.9 to 1578 pg/mL. Values obtained with the Nichols Advantage Intact PTH assay (y) were from 0.8 to 1502 pg/mL. A correlation coefficient (r) of 0.99 with a regression formula $y = 0.98x - 0.3$ was obtained from Deming linear regression analysis.

b. Matrix comparison:

Bias analysis and correlation was performed on three paired specimen types. Within serum (n=47), a small but detectable amount of bias was observed with the SST™ gel separator tube yielding on average 2.4 pg/mL higher results as compared to the plain Red Top tube. This amount of bias (+2.4 pg/mL) was considered to be within the known analytical variation of the test (see Intra and Inter-Assay Variation). For EDTA plasma specimens (n=48), no difference was observed in Intact PTH results between the two specimen collection tubes. For heparinized plasma specimens (n=45), no difference was observed in Intact PTH results between the two specimen collection tubes.

Intact PTH results, mean ±SD, for paired specimens: serum, EDTA plasma, and heparinized plasma.

	Serum (n=47)		EDTA plasma (n=48)		Heparinized plasma (n=45)	
Tube Type:	Red Top (plain)	SST™ gel separator	Lavender top EDTA	PPT™ gel separator	Green top heparin	PST™ gel separator
Mean ± SD (pg/mL)	44.1 ± 19.1	46.5 ± 19.8	41.1 ± 17.7	41.1 ± 18.7	40.4 ± 18.2	41.1 ± 18.9

Between Specimen Type Evaluation:

The same samples as described above were also evaluated for similarities and differences between specimen types. The data demonstrates the degree of correlation between specimen types. After paired t-testing, statistical difference between specimen types was observed. The minimum and maximum bias was considered within the expected analytical variation of the test for repeat measurements. There are detectable differences between specimen types for Intact PTH testing, however, these differences are small and are not considered different based upon the known analytical variation of the test.

Descriptive statistics comparing similarities and differences between specimen types for Intact PTH testing. Specimens were the same as described above.

Specimen Comparison	n	Range (pg/mL)	Paired *t-Test*	Bias Analysis			Deming Regression (r)
				Mean (pg/mL)	Min. (pg/mL)	Max. (pg/mL)	
Serum (x) vs. EDTA (y)	48	12.1-106.2 (Serum)	P < 0.001	3.6	-0.1	8.9	Y = 0.94x – 0.9 (r=0.99)
Serum (x) vs. Heparin (y)	48	11.3-101.3 (Heparin)	P < 0.001	2.7	-4.0	7.8	Y = 0.96x – 1.1 (r=0.99)
EDTA (x) vs. Heparin (y)	48	10.5-102.2 (EDTA)	P = 0.02	-0.9	-10.3	3.1	Y = 1.03x – 0.2 (r=0.99)

3. Clinical studies:
   - *a. Clinical sensitivity:*
     NA
   - *b. Clinical specificity:*
     NA
   - *c. Other clinical supportive data (when a and b are not applicable):*
     - Stability studies were performed on different tubes under different conditions, using the Nichols Advantage® Chemiluminescence Intact Parathyroid Hormone Immunoassay.

Stability of Intact PTH in EDTA anticoagulated whole blood when handled initially at room temperature (RT, 15-25°C) then refrigerated (2-8°C). The table shows percent recoveries (mean ± SD) after repeated processing of EDTA anticoagulated whole blood at 2 (baseline), 8, 24, and 48 hours after venipuncture. Specimens were assayed immediately after separation.

	2 hrs (baseline) RT	8 hrs 2-8°C	24 hrs 2-8°C	48 hrs 2-8°C
EDTA whole blood (dialysis specimens) n=33	100%	101 ± 6.0%	99 ± 3.0%	99 ± 4.0%

Plasma Intact PTH percent recoveries (mean ± SD) handled under refrigerated (2-8°C) conditions. Plasma specimens were processed within 2-hours of venipuncture, then the separated plasma was placed in the refrigerator and then frozen at –20°C at each time point. All time points for each specimen type were assayed within a single run.

Specimen Type	n	2 hrs (baseline) 2-8°C	8 hrs 2-8°C	24 hrs 2-8°C	48 hrs 2-8°C
Lavender Top tube: EDTA plasma	50	100.7 ± 3.0%	98.0 ± 4.5%	98.6 ± 4.6%	99.5 ± 4.6%
PPT™	50	99.5 ± 3.2%	97.0 ± 5.0%	96.9 ± 6.4%	98.1 ± 5.6%

EDTA plasma					
Green Top tube: Heparinized plasma	50	99.9 ± 2.9%	96.5 ± 5.1%	97.5 ± 4.6%	97.0 ± 4.3%
PST™ tube: Heparinized plasma	50	100.8 ± 3.7%	97.0 ± 5.0%	98.6 ± 5.9%	98.4 ± 4.5%

Plasma Intact PTH percent recoveries (mean ± SD) handled under room temperature (RT, 15-25°C) conditions. Plasma specimens were processed within 2-hours of venipuncture, the separated plasma was held at room temperature (15-25°C) and then frozen at –20°C at each time point. All time points for each specimen type were assayed within a single run.

Specimen Type	n	2 hrs (baseline) RT	8 hrs RT	24 hrs RT	48 hrs RT
Lavender Top tube: EDTA plasma	50	100.7 ± 3.0%	96.7 ± 3.8%	94.4 ± 4.2%	92.9 ± 5.2%
PPT™ EDTA plasma	50	99.5 ± 3.2%	96.2 ± 4.3%	96.5 ± 5.0%	95.2 ± 5.7%
Green Top tube: Heparinized plasma	50	99.9 ± 2.9%	98.1 ± 4.6%	96.2 ± 4.0%	97.3 ± 1.6%
PST™ tube: Heparinized plasma	50	100.8 ± 3.7%	97.1 ± 5.6%	97.4 ± 5.0%	92.9 ± 7.1%

The following specimen collection, preparation and storage instructions were established based on the results of the studies and are included in the labeling:

EDTA whole blood (with or without gel separator) can be handled and stored at refrigerated conditions (2-8°C) before centrifugation to separate plasma from cells. Alternately, EDTA whole blood may be centrifuged promptly after venipuncture and the separated plasma can be stored at refrigerated (2-8°C) or room temperature conditions, 15-25°C. Similarly, heparinized blood should be centrifuged promptly after venipuncture and the separated plasma can be stored at refrigerated or room temperature conditions. In all these instances, plasma Intact PTH testing should be completed within 48 hours after venipuncture. EDTA whole blood can be centrifuged immediately before testing or any time within 48 hours of venipuncture, within which time testing should be completed. The combined exposure of the sample either as whole blood or separated plasma should not exceed 48 hours. Sera should be rapidly processed and frozen within 2 hours of collection. Sera should be thawed rapidly and tested promptly. In addition, separating and freezing EDTA or heparinized

plasma (-20°C or colder) is also an acceptable method for transporting samples.

- The sponsor also provides references to support the claim which has been added to the Indications for Use that "Measurements of intact parathyroid hormone levels are also used as an aid in monitoring therapeutic intervention of secondary hyperparathyroidism that frequently occurs in chronic kidney disease."

4. Clinical cut-off:
   NA
5. Expected values/Reference range:
   Previously established for K962598. Normal range: 10 - 65 pg/mL with calcium values ranging from 8.8 - 10.5 mg/dL; patients with hypercalcemia of malignancy: from undetectable to 22 pg/mL with calcium values ranging from 10.5 - 17.6 mg/dL; patients with hypoparathyroidism: from undetectable to 21 pg/mL with calcium values ranging from 6.3 - 8.5 mg/dL. Because of the interplay between calcium and PTH levels in various parathyroid/calcium metabolism disorders, interpretation of PTH results should take the serum calcium concentrations into account.

N. Conclusion:

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