



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

I Background Information:

A 510(k) Number

K260331

B Applicant

Siemens Healthcare Diagnostics Products, Ltd.

C Proprietary and Established Names

IMMULITE 2000 3gAllergy Specific IgE Vantage;
IMMULITE 2000 Systems Peanut Allergen (F13)

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
DHB	Class II	21 CFR 866.5750 Radioallergosorbent (RAST) immunological test system	IM - Immunology

II Submission/Device Overview:

A Purpose for Submission::

New Device

B Measurand:

Allergen-specific Immunoglobulin E (IgE) to F13, Peanut

C Type of Test:

Quantitative, Chemiluminescent Immunoassay

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

The IMMULITE 2000 3gAllergy Specific IgE Vantage assay is for in vitro diagnostic use with the IMMULITE 2000 Systems Analyzers — for the quantitative measurement of allergen-specific IgE in human serum, as an aid in the clinical diagnosis of IgE-mediated allergic disorders. The test results are to be used in conjunction with clinical findings and other laboratory tests.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

D Special Instrument Requirements:

IMMULITE 2000 XPi System

IV Device/System Characteristics:

A Device Description:

The following materials are included in the IMMULITE 2000 3gAllergy Specific IgE Vantage:

- 3gAllergy Specific IgE Vantage Bead Pack, coated with anti-ligand
- 3gAllergy Specific IgE Vantage Accessory Reagent, monoclonal sheep anti-free biotin antibody in human serum albumin matrix, sodium azide, preservatives
- 3gAllergy Specific IgE Vantage Reagent Wedge, alkaline phosphatase (bovine calf intestine) conjugated to monoclonal murine anti-human IgE antibody in human/nonhuman serum buffer matrix
- 3gAllergy Specific IgE Adjustors, low and high levels of human IgE in nonhuman serum matrix, sodium azide, preservative
- 3gAllergy Specific IgE Vantage Adjustor Antibody, liquid, ready-to-use ligand-labeled polyclonal goat antihuman IgE antibody, preservative
- 3gAllergy Specific IgE (SPE) Universal Kit Controls, low and high levels of human IgE in a nonhuman serum matrix, sodium azide, preservative
- 3gAllergy Specific IgE Vantage Control Antibody, liquid, ready-to-use ligand-labeled polyclonal goat anti-human IgE antibody, preservative
- Specific Allergens, specific allergens in a protein-based buffer matrix, sodium azide preservative

Peanut (F13) is Specific Allergen in IMMULITE 2000 Systems Peanut Allergen (F13)

B Principle of Operation:

This assay is a solid-phase, 2-step chemiluminescent immunoassay that exploits liquid phase kinetics in a bead format. The solid phase (bead) is coated with anti-ligand. The liquid phase consists of alkaline phosphatase (bovine calf intestine) conjugated to monoclonal murine anti-human IgE antibody in a human/nonhuman serum buffer matrix. In the first cycle, the patient sample and ligand-labeled specific allergen and accessory reagent are incubated together with the coated bead for 30 minutes. During this time, specific IgE in the sample binds to the ligand-labeled allergen, which in turn binds to the anti-ligand on the bead. Unbound material is then removed by centrifugal washes. In the second cycle, the enzyme conjugated monoclonal murine anti-human IgE antibody is added to the original reaction tube for an additional 30-minute incubation. The enzyme conjugated monoclonal murine anti-human IgE antibody binds to immobilized IgE. The unbound enzyme conjugate is removed by centrifugal washes. Finally, chemiluminescent substrate is added to the reaction tube containing the bead and the signal is generated in proportion to the bound enzyme.

V Substantial Equivalence Information:**A Predicate Device Name(s):**

IMMULITE 2000 Allergen Specific IgE Kit

B Predicate 510(k) Number(s):

K013134

C Comparison with Predicate(s):

Device & Predicate	<u>K260331</u> (Candidate)	<u>K013134</u> (Predicate)
Device Trade Name	IMMULITE 2000 3gAllergy Specific IgE Vantage; IMMULITE 2000 Systems Peanut Allergen (F13)	IMMULITE 2000 Allergen Specific IgE Kit
General Device Characteristic Similarities		
Intended Use/ Indications For Use	The IMMULITE 2000 3gAllergy Specific IgE Vantage assay is for in vitro diagnostic use with the IMMULITE 2000 Systems Analyzers — for the quantitative measurement of allergen-specific IgE in human serum, as an aid in the clinical diagnosis of IgE-mediated allergic disorders. The test results are to be used in conjunction with clinical findings and other laboratory tests.	The IMMULITE 2000 Allergen Specific IgE is for in vitro diagnostic use with the IMMULITE 2000 Automated Immunoassay Analyzer – for the measurement of allergen-specific IgE in human serum, as an aid in the clinical diagnosis of IgE-mediated allergic disorders.

Device & Predicate	<u>K260331</u> (Candidate)	<u>K013134</u> (Predicate)
Technology	Chemiluminescent	Same
Traceability	Traceable to the WHO 2 nd International Reference Preparation (75/502)	Same
Sample type	Serum	Same
Controls	Specific IgE Control Antibody and Specific IgE Universal Controls	Same
Process Time	65 minutes	Same
Detection Antibody	Monoclonal murine anti-human IgE antibody conjugated to alkaline phosphatase in a human/nonhuman serum buffer matrix	Same
Kit bead pack (solid phase)	Polystyrene bead coated with biotinylated methylated human serum albumin and streptavidin.	Same
General Device Characteristic Differences		
Allergen	Whole Peanut Extract (F13)	Foods (F), Grass pollen (G), Weed pollen (W), Tree pollen (T), Epidermal and animal proteins (E), Microorganisms (M), Mites (D), Venoms and insects (I), Occupational (K)
Accessory Reagent	Recombinant sheep monoclonal antibody high biotin affinity reagent	None
Sample	30 µL	50 µL

VI Standards/Guidance Documents Referenced:

The following Clinical and Laboratory Standards Institute (CLSI) guidelines and International Organization for Standardization (ISO) were used:

- CLSI EP05-A3, Evaluation of Precision of Quantitative Measurement Procedures; Approved Guideline—Third Edition
- CLSI EP06 2nd Edition, Evaluation of 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 EP25 2nd Edition, Evaluation of Stability of In Vitro Medical Laboratory Test Reagents

- 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
- CLSI EP39 1st Edition, A Hierarchical Approach to Selecting Surrogate Samples for the Evaluation of In Vitro Medical Laboratory Tests
- CLSI I/LA20 3rd Edition, Analytical Performance Characteristics, Quality Assurance, and Clinical Utility of Immunological Assays for Human Immunoglobulin E Antibodies of Defined Allergen Specificities
- ISO 17511:2020, In vitro diagnostic medical devices — Requirements for establishing metrological traceability of values assigned to calibrators, trueness control materials and human samples—Second Edition

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

The studies were performed in accordance with CLSI guideline EP05-A3.

Within Laboratory precision:

The study was performed at a single site using one lot of the IMMULITE 2000 Systems Peanut Allergen reagent on one IMMULITE 2000 XPi instrument. Five levels of human serum samples were run in two replicates per run, two runs daily over the course of 21 days, resulting in a minimum of 84 datapoints for each sample. The data were analyzed for repeatability (within-run), between-run, between-day, and within-laboratory precision. The mean (kU/L), standard deviation (SD) (kU/L) and percent coefficient of variation (%CV) are summarized in table below.

Sample	N*	Mean (kU/L)	Within-Run (Repeatability)		Between-Run		Between-Day		Within-Laboratory	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
1	84	0.19	0.02	8.6	0.01	5.2	0.01	4.8	0.02	11.2
2	83	0.26	0.01	5.1	0.01	4.5	0.00	1.4	0.02	6.9
3	83	0.43	0.02	4.9	0.00	0.0	0.02	3.8	0.03	6.2
4	84	1.48	0.06	3.8	0.06	3.8	0.02	1.1	0.08	5.5
5	84	83.20	3.19	3.8	1.23	1.5	1.00	1.2	3.56	4.3

*Sample #2 and #3 each has one replicate excluded from analysis.

Lot-to-Lot Precision:

The study was performed was performed using three lots of IMMULITE 2000 Systems Peanut Allergen reagent. Seven levels of serum samples were run in five replicates per run, one run per day over the course of five days, resulting in a total of 75 datapoints for each

sample. The data were analyzed for repeatability (within-run), between-day, between-reagent lot, and total precision. The mean (kU/L), standard deviation (SD) (kU/L) and percent coefficient of variation (%CV) are summarized in table below:

Sample	N	Mean (kU/L)	Within-Run (Repeatability)		Between-Run		Between-Lot		Total	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
1	75	0.17	0.02	12.2	0.00	0.0	0.01	6.5	0.02	13.8
2	75	0.27	0.02	6.3	0.01	3.9	0.00	0.0	0.02	7.4
3	75	0.65	0.04	5.7	0.01	1.5	0.06	8.5	0.07	10.4
4	75	1.42	0.07	5.2	0.04	2.9	0.12	8.5	0.15	10.3
5	75	4.88	0.19	4.0	0.19	4.0	0.36	7.3	0.45	9.2
6	75	25.10	1.27	5.1	0.00	0.0	1.30	5.2	1.81	7.2
7	75	69.40	3.39	4.9	1.87	2.7	5.86	8.4	7.03	10.1

Instrument-to-instrument precision:

The study was performed using three IMMULITE 2000 XPi instruments with a single lot of the IMMULITE 2000 Systems Peanut Allergen reagent. Five levels of human serum samples were run in five replicates per run, one run per day over the course of five days, resulting in a total of 75 datapoints for each sample. The data was analyzed for repeatability (within-run), between-day, between-instrument, and reproducibility. The mean (kU/L), standard deviation (SD) (kU/L) and percent coefficient of variation (%CV) are summarized in the table below:

Sample	N	Mean (kU/L)	Within-Run (Repeatability)		Between-Day		Between-Instrument		Total	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
1	75	0.15	0.02	11.6	0.00	0.0	0.01	3.5	0.02	12.1
2	75	0.28	0.03	9.1	0.01	2.8	0.02	5.5	0.03	11.0
3	75	0.60	0.03	5.3	0.00	0.0	0.01	1.1	0.03	5.5
4	75	1.28	0.07	5.0	0.04	3.1	0.02	1.9	0.08	6.2
5	75	78.90	4.17	5.3	2.73	3.5	0.00	0.0	4.99	6.3

2. Linearity:

The linearity of the IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen was performed in accordance with CLSI guideline EP06, 2nd Edition. A fourteen (14) level dilution series was prepared by mixing the high serum samples and low levels serum samples in known ratios. The dilution levels were run in a minimum of three replicates on one IMMULITE 2000 XPi instrument using one reagent lot. The 'measured value' (mean kU/L) was compared to its predicted value for percentage of deviation using a linear regression analysis. This deviation was then compared to the allowable deviation from linearity (ADL). The regression results are summarized in the table below.

Range (kU/L)	Slope (95% CI)	Intercept (95% CI)	R ²
0.061 - 131	1.03 (1.00 - 1.06)	0*	0.996

*95% CI is not applicable

The study results support the linearity of the claimed analytical measuring interval (AMI) for IMMULITE 2000 Systems Peanut Allergen: 0.100–100 kU/L.

3. Analytical Specificity/Interference:

Inhibition study:

Inhibition study was performed according to CLSI guidelines I/LA20 (3rd Edition) to determine the analytical specificity of the IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen by competitive inhibition. One related allergen (un-biotinylated F13) and three unrelated allergens, including D1 (Dermatophagoides Pteronyssinus), E2 (Dog Epithelium), and M6 (Alternaria Tenuis) were tested. The related allergen was diluted to create five dilutions: 1:5, 1:25, 1:125, 1:625, and 1:312, each evaluated for dose-dependent inhibition. The unrelated allergens were prepared and tested at a single concentration that is at least 10 folds higher than that of the related allergen at which greater than 85% inhibition was achieved. The related or unrelated allergen was spiked 1:1 into aliquots of a positive Peanut Allergen human serum sample (10.4 kU/L). Each sample was assayed in duplicate using one reagent lot on one IMMULITE 2000 XPi instrument. The results of the inhibition studies for the related allergen (F13) showed 82%, 92% and 97% inhibition at a concentration of 4, 20 and 100 µg/mL, respectively. The inhibition studies, using unrelated allergens at a test concentration of 100 µg/mL did not show any significant inhibition. The inhibition studies indicate that the IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen contains the immunologically relevant allergen.

Interference:

Interference study was performed according to CLSI guidelines EP07, 3rd Edition and EP37, 1st Edition to determine the effect of various potential endogenous and exogenous interfering substances on the IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen. Two serum sample pools with target specific IgE concentrations of approximately 0.300–0.699 kU/L and 0.700–3.50 kU/L were spiked with and without the interferent and measured in seven replicates using one reagent lot on a single IMMULITE 2000 XPi instrument. The percent difference was determined by comparing the result of the sample spiked with interferent to that of control sample without the interferent. The percent difference between the mean spiked sample value and the mean control value was calculated $[100 * (\text{test mean} - \text{control mean}) / \text{control mean}]$ and an interference within $\leq 10\%$ difference was considered non-significant.

a) Endogenous Substance Interference:

The following potential endogenous interfering substances were tested using IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen. No significant interference was found for each potential interfering substance at the concentrations listed below.

Endogenous Substance	Concentration
Albumin	120 g/L
Bilirubin (conjugated)	40 mg/dL
Bilirubin (unconjugated)	40 mg/dL
Hemoglobin	1,000 mg/dL
Intralipid	3,000 mg/dL
Rheumatoid Factor	500 IU/mL
HAMA* Type I	87 ng/mL
HAMA Type II	107 ng/mL

* human anti-mouse antibodies

b) Exogenous Substance Interference:

The following potential exogenous interfering substances were tested using IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen. No significant interference was found each potential interfering substance at the concentrations listed below.

Exogenous Substance	Concentration
diphenhydramine	19.5 µmol/L
methylprednisolone	7,830 ng/mL
ranitidine	33.4 µmol/L
omalizumab	0.12 mg/mL

Biotin interference: Significant interference was found for biotin at high concentrations as listed in the table below.

Sample (kU/L)	% Bias				
	Biotin concentration (ng/mL)				
	500	600	1,000	1,500	3,510
0.44	-6.5%	0.5%	-5.4%	-10.8%	-77.5%
1.3	0.8%	-0.8%	-6.2%	-5.4%	-92.3%

The IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen ‘instructions for use’ contains the following statement of limitation “*testing specimens from patients taking biotin may lead to falsely depressed results. Therefore, do not use this device in patients with IgE-mediated allergic disorders, unless it is confirmed that the patient is not taking high doses of biotin.*”

Cross-reactivity:

The potential cross-reactivity of the following non-IgE (IgA, IgD, IgG, and IgM) was evaluated using IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen according to the CLSI guideline EP07, 3rd Edition. No significant cross-reactivity (percent difference ≤ 10%) was found for each cross-reactant tested at the concentration listed below.

Cross-Reactant	Concentration
Immunoglobulin G	2000 mg/dL
Immunoglobulin A	500 mg/dL
Immunoglobulin M	400 mg/dL
Immunoglobulin D	240 mg/L

4. Detection Capability:

CLSI guideline EP17-A2 was followed to determine the Limit of Blank (LoB), Limit of Detection (LoD) and Limit of Quantitation (LoQ) for the IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen.

The LoB was determined by testing four analyte undetectable serum samples over five days, one runs per day, five replicates per run using two reagent lots on one IMMULITE 2000 XPi instrument for a total of 100 replicates per reagent lot. The LoB for each reagent lot was calculated using the non-parametrically at the 95th percentile of all measurements of the blank samples. The LoB was calculated separately for each reagent lot, and the highest value was taken as the LoB value. The LoB values from the two lots were all 0.0 kU/L. The claimed LoB is 0.0 kU/L.

The LoD was determined by testing four serum samples diluted with serum samples containing low levels of measurand were tested over five days, one run per day, five replicates per run using two reagent lots on IMMULITE 2000 XPi instrument for a total of 100 replicates per reagent lot. The LoD was analyzed by using a parametric method approach. The LoD was calculated separately for each reagent lot, and the highest value was taken as the LoD value. The LoQ values from the two lots were 0.036 kU/L and 0.044 kU/L. The claimed LoD for IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen on the IMMULITE 2000 XPi System is 0.044 kU/L.

The LoQ was determined by testing nine serum samples diluted with serum samples containing low levels of measurand over five days, two runs per day, five replicates per run using two reagent lots on one IMMULITE 2000 XPi instrument for a total of 50 replicates per sample per reagent lot. For each reagent lot, the within-laboratory precision over five days, expressed as %CV, was plotted against the mean measurand concentration for each sample. The LoQ was defined as the level at which the %CV of the within-laboratory imprecision of no more than 20%. The LoQ values from the two lots were 0.052 kU/L and 0.079 kU/L. The claimed LoQ for the IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen results on the IMMULITE 2000 XPi System is 0.1 kU/L.

5. Assay Reportable Range:

The analytical measuring interval (AMI) for the IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen on the IMMULITE 2000 XPi System is 0.100–100 kU/L. Values below the low limit of AMI are reported as <0.1 kU/L. Values above the upper limit of AMI are reported as > 100 kU/L.

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

Traceability:

The calibrators and controls are traceable to the World Health Organization (WHO) 2nd International Reference Preparation 75/502 reference standard for human IgE.

Kit Stability:

The stability of the IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen reagent kit was determined according to CLSI guideline EP25, 2nd Edition.

a) Shelf-life stability:

A real-time stability study for the IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen stored at 2–8°C is on-going by testing four samples (one negative and three positive samples) at Day 0 and at scheduled timepoints of 1, 2, 7, 30, 62, 120, 183, 273, 330, 366, and 391 days in four replicates per sample using three reagent kit lots on one IMMULITE 2000 XPi instrument. The current data support stability of the IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen for 12 months at 2–8°C.

b) On-board and Open vial stability

An on-board and accessory reagent open vial stability study was performed on one IMMULITE 2000 XPi instrument using four samples (one negative and three positive samples) and one lot of IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen. The testing was performed for each sample in four replicates at Day 0 and scheduled timepoints of 1, 2, 7, 14, 28, 56, 84, 87, 90, 91 days (in-use) and fresh reagent lots were used to assay each sample. The accessory reagent was aliquoted and left uncapped for ≥ 7 days at room temperature (15–28°C) to simulate aging. The data supports stability of the IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen onboard and accessory reagent open vial stability for 90 days.

7. Assay Cut-Off:

Not applicable

B Comparison Studies:

1. Method Comparison with Predicate Device:

CLSI guidelines EP09c 3rd Edition and I/LA20 3rd Edition were followed to compare the IMMULITE 2000 3gAllergy Specific IgE Vantage with Peanut Allergen to the IMMULITE 2000 3gAllergy Specific IgE Universal Kit with Peanut Allergen. A total of 87 serum samples with specific IgE concentrations falling within the AMI (0.16–81.3 kU/L) were tested across

three days with one reagent lot of IMMULITE 2000 3gAllergy Specific IgE Vantage assay with Peanut Allergen (candidate) and one reagent lot of IMMULITE 2000 3gAllergy Specific IgE Universal Kit with Peanut Allergen (predicate) on the IMMULITE 2000 XPi instrument. The comparison between paired measurements was analyzed using Passing-Bablok method. The results are summarized in the table below.

N	Range (kU/L)*	Slope (95% CI)	Intercept (95% CI)	R
87	0.16–81.3	0.95 (0.92, 1.00)	-0.089 (-0.141, -0.047)	0.99

2. Matrix Comparison:

Not applicable

C Clinical Studies:

1. Clinical Sensitivity and Specificity

The clinical performance was evaluated by comparing the test results from a study cohort including 132 atopic peanut-allergic individuals (subjects with a clinical history of symptoms consistent with an IgE-mediated food allergic reaction upon exposure to peanut as diagnosed by a physician) and 85 atopic with no peanut allergy individuals (participants having no IgE-mediated allergic responses to peanut, and having clinical history of IgE-mediated allergic responses to foods other than peanut or to environmental allergens as diagnosed by a physician). The IMMULITE 2000 3gAllergy Specific IgE Vantage assay results are expressed as positive or negative using a cut-off of 0.35 kU/L. The clinical sensitivity and clinical specificity, with 95% confidence intervals (CI), for IMMULITE 2000 Specific IgE Vantage assay with Peanut Allergen assay are summarized in the following table:

		Clinical Diagnosis		
		Atopic Peanut	Atopic Non-Peanut	Total
IMMULITE 2000 Specific IgE Vantage, Peanut Allergen	Positive (≥ 0.35 kU/L)	110	5	115
	Negative (< 0.35 kU/L)	22	80	102
	Total	132	85	217
Clinical Sensitivity: 83% (110/132) (95% CI: 76 – 89%)				
Clinical Specificity: 94% (80/85) (95% CI: 87 – 97%)				

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

Not Applicable

3. Clinical Cut-Off:

The clinical cut-off of the assay is 0.35 kU/L. The interpretation of results of the assay uses the following classification system:

Class	Concentration (kU/L)	Interpretation
0*	<0.35	Negative
I	0.35 to 0.69	Positive with increasing sIgE concentration
II	0.70 to 3.49	
III	3.50 to 17.49	
IV	17.50 to 52.49	
V	52.50 to 99.99	
VI	≥100	

* Class 0 in the standard system signifies not detectable by second-generation assays

D Expected Values/Reference Range:

The reference interval for the IMMULITE 2000 3gAllergy Specific IgE Vantage for Peanut Allergen (F13) was determined in accordance with CLSI EP28-A3c based on the test results using 108 apparently healthy (non-atopic) individuals. All samples were tested below 0.35 kU/L.

Each laboratory should establish its own expected value/reference range.

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.