

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

A. 510(k) Number:

k093987

B. Purpose for Submission:

Modification to device

C. Measurand:

Twenty one new Individual Allergen Specific IgE: Aureobasidium pullulans, Blomia tropicalis, Brazilian Peppertree, Budgerigar Feathers, Canary feathers, Chicken Feathers, Loblolly Pine, nAsp r 1, nCan f 1, nDer f 1, nDer f 2, nDer p 1, nDer p 2, nFel d 1, nPen m 1, Parrot feathers, Penicillium brevi-compactum, Pigeon Droppings, Rabbit bush, Tobacco, Turkey feathers

D. Type of Test:

Quantitative, chemiluminiscent immunoassay

E. Applicant:

Siemens Healthcare Diagnostics, Inc.

F. Proprietary and Established Names:

IMMULITE® 2000 3gAllergy™ Specific IgE Assay kit

G. Regulatory Information:

1. Regulation section:
21 CFR § 866.5750, Radioallergosorbent (RAST) immunological test system
2. Classification:
Class II
3. Product code:
DHB System, Test, Radioallergosorbent (RAST), Immunological
4. Panel:
Immunology (82)

H. Intended Use:

1. Intended use:
For *in vitro* diagnostic use with the IMMULITE 2000 Analyzer – 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.
2. Indication(s) for use:
Same as Intended use.
3. Special conditions for use statement(s):
For prescription only.
4. Special instrument requirements:
IMMULITE 2000 Analyzer (k970227)

I. Device Description:

Each device contains the following: 3gAllergy™ specific IgE bead pack (3 packs of 200 beads coated with anti-ligand); specific IgE reagent wedge: 30 mL alkaline phosphatase (bovine calf intestine) conjugated to monoclonal murine anti-human IgE antibody in a human/nonhuman serum buffer matrix (equally dispensed in 1 wedge

with B & C chambers); specific IgE adjustors: low and high (2 vials, 2 mL each) of human IgE in a nonhuman serum matrix with preservative; specific IgE adjustor antibody: 2 tubes, 2.75 mL each) ready to use ligand-labeled polyclonal goat anti-human IgE antibody with preservative; specific IgE universal kit controls: (2 vials, 2 mL each) human IgE in a nonhuman sample matrix with preservative; specific IgE control antibody: (2 tubes, 2.75 mL each) ready to use ligand-labeled polyclonal goat anti-human IgE antibody with preservative. Kit components supplied separately: 3gAllergy™ specific IgE sample diluent (concentrated ready to use 1 vial, 25 mL); chemiluminiscent substrate; probe wash; probe cleaning kit; disposable reaction tubes; bar coded allergen holder wedges serially coded 1-33; 34 -66; 67-99; allergen tube caps and tube septa.

J. Substantial Equivalence Information:

1. Predicate device name(s):
IMMULITE® 2000 3gAllergy™ Specific IgE
2. Predicate K number(s):
k013134
3. Comparison with predicates:

Similarities		
Item	New Device	Predicate Device
Intended use	For <i>in vitro</i> diagnostic use with the IMMULITE 2000 Analyzer – 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.	Same
Technology	Chemiluminescence	Same
Assay performance	Specific to allergen-specific IgE	Same
Calibrators	Low and high	Same
Controls	Specific IgE and Antibody and Specific IgE Universal Controls	Same
Sample type	Serum	Same
Result Interpretation	Quantitative values in kU/L; Interpretation of class results for two scoring systems: Standard and Extended standard	Same

Differences		
Item	New Device	Predicate Device
Total number of Allergens	Twenty one	One hundred and ten
Types of	21 Specific Allergens:	110 Specific Allergens:

Differences		
Item	New Device	Predicate Device
Specific Allergens	9 Animal epithelia; 5 Mites; 1 Weed; 3 Molds/Fungi; 2 Trees; 1 Miscellaneous (Tobacco)	8 Animal epithelia, 2 House dusts, 39 Food, 13 Grasses, 5 Insects, 5 Mites, 10 Molds, 1 Latex, 14 Trees, 13 Weeds

K. Standard/Guidance Document Referenced (if applicable):

FDA Guidance – Radioallergosorbent Test (RAST) Methods for Allergen-Specific Immunoglobulin E (IgE) 510(k); Final Guidance

CLSI I/LA 20-A: Evaluation Methods and Analytical Performance Characteristics of Immunological Assays for Human Immunoglobulin E (IgE)

CLSI EP5-A2: Evaluation of Precision Performance of Quantitative Methods; Approved Guideline – Second Edition

L. Test Principle:

The assay is a solid-phase, two-step, chemiluminiscent immunoassay that exploits liquid phase kinetics in a bead format. The allergens are covalently bound to a soluble polymer/co-polymer matrix, which is labeled with a ligand. The assay specific antibody is labeled with alkaline phosphatase. The use of an amino acid co-polymer amplifies the amount of allergen that the matrix can support. The chemiluminiscent detection system is a phosphatase ester of stabilized dioxatane. Cleavage of the phosphate ester by alkaline phosphatase results in the decomposition of dioxatane and the emission of photons, which are quantified by a luminometer.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

Reproducibility of the assay was assessed by testing three positive samples and one negative control sample of each allergen (Aureobasidium pullulans, Blomia tropicalis, Brazilian Peppertree, Budgerigar Feathers, Canary feathers, Chicken Feathers, Loblolly Pine, nAsp r 1, nCan f 1, nDer f 1, nDer f 2, nDer p 1, nDer p 2, nFel d 1, nPen m 1, Parrot feathers, Penicillium brevicompactum, Pigeon Droppings, Rabbit bush, Tobacco, Turkey feathers) in duplicate twice a day for 20 different days (n = 80). The sponsor's criterion for the negative sample was the average dose level must be <0.10 kU/L; all negative sample mean results were within the acceptance criterion. The sponsor's acceptance criterion for the positive samples was ≤15% CV for both within-run and total precision. Three allergen lots were tested for each allergen; representative data from one lot is shown below for the positive samples. Additional studies for Positives #4 were done assaying two aliquots of each test sample in two runs per day on ten different days. Analysis of variance was used to estimate the within and total precision. The intra-assay and inter-assay %CV ranges were from 2.6% to 13.28 % and 3.59% to 14.90%, respectively (see tables below).

Allergen: Aureobasidium pullulans

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	0.72	0.027	3.75	0.041	5.69
Positive #2	0.96	0.037	3.85	0.041	4.27
Positive #3	12.19	0.529	4.34	0.788	6.46
Positive #4	0.54	0.072	13.28	0.081	14.90

Allergen: Blomia tropicalis

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	36.21	1.403	3.87	1.645	4.54
Positive #2	1.73	0.133	7.69	0.169	9.77
Positive #3	2.55	0.079	3.10	0.121	4.75
Positive #4	0.42	0.023	5.36	0.031	7.20

Allergen: Brazilian Peppertree

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	6.03	0.179	2.97	0.282	4.68
Positive #2	53.50	1.456	2.72	1.922	3.59
Positive #3	1.52	0.070	4.61	0.079	5.20

Allergen: Budgerigar Feathers

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	0.45	0.026	5.78	0.029	6.44
Positive #2	1.35	0.070	5.19	0.095	7.04
Positive #3	1.89	0.094	4.07	0.159	8.41
Positive #4	4.34	0.144	3.32	0.165	3.81

Allergen: Canary feathers

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	0.74	0.028	3.78	0.038	5.14
Positive #2	0.99	0.033	3.33	0.048	4.85
Positive #3	6.80	0.271	3.99	0.312	4.59
Positive #4	0.41	0.050	3.76	0.027	6.71

Allergen: Chicken Feathers

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	20.40	0.771	3.78	1.778	8.72
Positive #2	4.98	0.186	3.73	0.645	12.95
Positive #3	1.00	0.026	2.60	0.071	7.10

Allergen: Loblolly Pine

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	1.05	0.036	3.43	0.052	4.95
Positive #2	0.87	0.051	5.86	0.055	6.32
Positive #3	6.00	0.295	4.92	0.381	6.35
Positive #4	0.31	0.016	5.25	0.021	6.71

Allergen: nAsp r 1

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	1.78	0.082	4.61	0.111	6.24
Positive #2	62.51	2.541	4.06	3.704	5.93
Positive #3	0.46	0.026	5.65	0.033	7.17

Allergen: nCan f 1

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	3.29	0.122	3.71	0.178	5.41
Positive #2	9.03	0.386	4.27	0.503	5.57
Positive #3	21.90	0.796	3.63	0.887	4.05

Allergen: nDer f 1

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	3.22	0.142	4.41	0.179	5.56
Positive #2	9.16	0.394	4.30	0.486	5.31
Positive #3	65.82	2.478	3.76	3.046	4.63

Allergen: nDer f 2

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	71.90	2.835	3.94	3.325	4.62

Positive #2	12.63	0.666	5.27	0.941	7.45
Positive #3	13.61	0.646	4.75	0.800	5.88
Positive #4	0.54	0.028	5.44	0.039	7.36

Allergen: nDer p 1

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	50.96	2.143	4.21	2.450	4.81
Positive #2	7.38	0.328	4.44	0.427	5.79
Positive #3	38.26	1.791	4.68	2.087	5.45
Positive #4	0.45	0.018	4.11	0.027	5.96

Allergen: nDer p 2

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	29.76	1.319	4.43	1.887	6.34
Positive #2	13.79	0.698	5.06	0.922	6.69
Positive #3	9.55	1.127	11.80	1.203	12.60
Positive #4	0.34	0.021	6.35	0.029	8.62

Allergen: nFel d 1

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	5.08	0.249	4.90	0.353	6.95
Positive #2	20.83	1.016	4.88	1.171	5.62
Positive #3	1.75	0.071	4.06	0.093	5.31

Allergen: nPen m 1

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	3.21	0.146	4.55	5.26	5.26
Positive #2	2.25	0.094	4.18	6.71	6.76
Positive #3	30.79	1.409	4.58	5.14	5.26
Positive #4	0.42	0.020	4.64	0.025	5.88

Allergen: Parrot feathers

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	1.14	0.056	4.91	0.073	6.40
Positive #2	0.32	0.020	6.25	0.025	7.81

Positive #3*	0.77	0.307	39.87	0.362	47.01
Positive #3**	0.72	0.038	5.28	0.091	12.64
Positive #4	10.74	0.461	4.29	0.025	5.88

Allergen: Penicillium brevi-compactum

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	1.87	0.109	5.83	0.127	6.79
Positive #2	1.37	0.053	3.87	0.070	5.11
Positive #3	4.87	0.190	3.90	0.226	4.64
Positive #4	0.43	0.013	3.05	0.018	4.15

Allergen: Pigeon Droppings

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	0.48	0.033	6.88	0.042	8.75
Positive #2	6.22	0.401	6.45	0.497	7.99
Positive #3	5.33	0.232	4.35	0.261	4.90
Positive #4	2.27	0.111	4.88	0.178	7.85

Allergen: Rabbit bush

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	2.48	0.098	3.95	0.142	5.73
Positive #2	3.61	0.124	3.43	0.205	5.68
Positive #3	52.20	1.571	3.01	2.321	4.45
Positive #4	0.46	0.017	3.69	0.029	6.26

Allergen: Tobacco

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	4.83	0.165	3.42	0.313	6.48
Positive #2	2.34	0.074	3.17	0.084	3.59
Positive #3	0.48	0.016	3.33	0.023	4.79

Allergen: Turkey feathers

Sample	Mean (kU/L)	Intra-assay		Inter-assay	
		SD (kU/L)	%CV	SD (kU/L)	%CV
Positive #1	0.54	0.021	3.89	0.027	5.00
Positive #2	0.75	0.027	3.60	0.035	4.67

Positive #3	30.23	1.004	3.32	1.345	4.45
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Additional precision studies were performed on thirteen allergens for the following concentration levels (see table below):

Class	Specific Allergen	Mean	With-in Run		Total	
			SD	CV	SD	CV
Class I	M12 Aureobasidium pullulans	0.54	0.072	13.28	0.081	14.90
0.35-0.69 kU/L						
Class I	E201 Canary Feathers	0.41	0.05	3.76	0.027	6.71
0.35-0.69 kU/L						
Class 0/I	T43 Loblolly Pine	0.31	0.016	5.25	0.021	6.71
0.10-0.69 kU/L						
Class I	A302 nDer f 2	0.54	0.028	5.44	0.039	7.36
0.35-0.69 kU/L						
Class I	A310 nDer p 1	0.45	0.018	4.11	0.027	5.96
0.35-0.69 kU/L						
Class 0/ I	A316 nDer p 2	0.34	0.021	6.35	0.029	8.62
0.10-0.69 kU/L						
Class I	F351 nPen m 1	0.42	0.020	4.64	0.025	5.88
0.35-0.69 kU/L						
Class I	M305 Penicillium brevi- compactum	0.43	0.013	3.05	0.018	4.15
0.35-0.69 kU/L						
Class I	W36 Rabbit Bush	0.46	0.017	3.69	0.029	6.26
0.35-0.69 kU/L						
Class II	D210 Blomia tropicalis	0.42	0.023	5.36	0.031	7.20
0.70 – 3.49 kU/L						
Class II	E7 Pigeon Droppings	2.27	0.111	4.88	0.178	7.85
0.70 – 3.49 kU/L						
Class III	E91 Parrot Feathers	10.74	0.461	4.29	0.025	5.88
3.50-17.49 kU/L						
Class III	E78 Budgerigar Feathers	4.34	0.144	3.32	0.165	3.81
3.50-17.49 kU/L						

Lot to lot imprecision:

The three tested lots were analyzed for lot-to-lot precision using three positive and one negative sample. Within run imprecision between lots ranged from 3.26% to 18.79 % and total imprecision from 4.36% to 18.79 %. All three lots were within the sponsor's claimed acceptable criterion of $\leq 20\%$ variability.

b. Linearity/assay reportable range:

Linearity studies: For each allergen, two clinical samples were diluted in 2-fold serial dilutions to 5 levels. The undiluted (neat) and diluted samples were tested with the specific allergen to demonstrate linearity at concentrations within the assay limits. Regression statistics for each allergen comparing the observed results to expected results are presented below:

Allergen	Regression Equation	N	Slope 95% CI	Intercept 95% CI
M12 – Aureobasidium Pullulans	$Y = 0.9964x - 0.0296$	9	0.968 – 1.025	-0.089 – 0.029
D201 – Blomia tropicalis	$Y = 0.9877x + 0.0284$	12	0.967 – 1.008	-0.063 – 0.119
T401 – Brazilian Peppertree	$Y = 0.9818x + 0.215$	12	0.939 – 1.024	0.070 – 0.360
E78 – Budgerigar Feathers	$Y = 0.939x + 0.224$	10	0.85 – 1.03	0.102 – 0.346
E201 – Canary Feathers	$Y = 1.012x + 0.2368$	11	0.907 – 1.117	-0.049 – 0.522
E85 – Chicken Feathers	$Y = 1.012x + 0.2814$	12	0.964 – 1.059	-0.079 – 0.642
T43 – Loblolly Pine	$Y = 1.009x - 0.1098$	12	0.994 – 1.023	-0.177 – 0.043
A3050 – nAsp r 1	$Y = 1.005x - 0.0923$	10	0.989 – 1.021	-0.299 – 0.114
A174 – nCan f 1	$Y = 0.993x + 0.0659$	12	0.985 – 1.001	0.037 – 0.095
A295 – nDer f 1	$Y = 1.002x + 0.1762$	12	0.979 – 1.024	-0.018 – 0.370
A302 – nDer f 2	$Y = 0.994x + 0.0236$	12	0.982 – 1.006	-0.038 – 0.085
A310 – nDer p 1	$Y = 0.996x - 0.0213$	12	0.978 – 1.104	-0.122 – 0.079
A316 – nDer p 2	$Y = 0.997x + 0.0978$	12	0.987 – 1.007	0.042 – 0.154
A345 – nFel d 1	$Y = 0.997x + 0.0484$	12	0.964 – 1.030	-0.203 – 0.299
F351 – nPen m 1	$Y = 1.002x + 0.1198$	11	0.982 – 1.022	0.009 – 0.231
E91 – Parrot Feathers	$Y = 0.992x + 0.4060$	12	0.919 – 1.065	0.066 – 0.746
M305 – Penicillium brevi-compactum	$Y = 0.979x + 0.2574$	12	0.864 – 1.093	-0.001 – 0.516
E7 – Pigeon Droppings	$Y = 0.959x + 0.0758$	10	0.893 – 1.025	0.0369 – 0.1147
W36 – Rabbit Bush	$Y = 0.982x + 0.4142$	12	0.914 – 1.050	0.150 – 0.679

Allergen	Regression Equation	N	Slope 95% CI	Intercept 95% CI
0201 - Tobacco	$Y = 0.964x + 0.2661$	12	0.916 – 1.012	0.139 – 0.393
E89 – Turkey Feathers	$Y = 0.993x + 0.2334$	12	0.932 – 1.053	0.058 – 0.409

IMMULITE 2000 3gAllergy Universal Kit Total IgE working ranges:
0.1 - 100 kU/L

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

Traceability: The calibrators and controls are traceable to the WHO 2nd IRP 75/502 reference standard for human IgE.

Stability:

Allergen stability: Accelerated allergen stability testing (15-25°C for 57 days; assay kits stored at recommended temperature 2-8°C). The accelerated study supports a two year shelf-life stability claim.

On-board/ open stability: Testings were performed on two positive samples and one negative sample on days 1, 7, 16, 21, 30, 45, 62, 76, 91 for each individual specific allergens. Stability studies support the 90 day stability claim.

Adjustment interval (calibration curve) stability: Testing was performed at days 1 and 14 to validate the 2-week adjustment interval. The calibration curve stability study supports the 2 weeks stability claim.

Sample stability: Stress conditions representing the storage claims were set up at 3 and 7 days at 2-8°C and 6 months at -20 °C. No significant variation to the reference samples that were run at day 0 was observed. The sample stability study supports the 7 days at 2-8°C and 6 months at -20 °C stability claims.

d. *Detection limit:*

Limit of Blank (LoB): Three runs assaying the blank sample (zero calibrator) were performed to estimate the LoB. A total of three instruments were used per run. The maximum dose of the LoB was selected as the most conservative LoB: $LoB_{MAX} = 0.026$. The claimed LoB is 0.1 kU/L.

Limit of Detection (LoD): Five samples were used to estimate the LoD. Sixty replicates of each sample were assayed per run. A total of 2 runs were completed with testing performed on 2 different instruments. The LoD was calculated for each sample using the formula: $LoD = LoB_{MAX} + (1.65 * SD_{LOD})$. The claimed LoD is 0.1 kU/L

e. *Analytical specificity:*

Inhibition studies: Specificity of each allergen was verified through competitive inhibition testing using a single serum sample or pool of sera. A negative sample was used to measure the background response.

To initiate the inhibition experiment, 70 µL of undiluted and 3-4 levels of 5-

fold serially diluted inhibitor extract were mixed with 250 µL of sample or pool to achieve final inhibitor concentrations of 218.75, 43.75, 8.75, 1.75, 0.35, 0.08, 0.07, 0.02, 0.01, 0.003 µg/mL inhibitor. This mixture was incubated at room temperature (15-28 °C) for 1 hour allowing the immunological reaction to occur. Each sample mixture containing the inhibitor extract and the appropriate controls was assayed with 1 lot of each allergen. The percent (%) inhibition was calculated according to the following formula:

$$\frac{(\text{Response of pos. control} - (\text{pos. sample} - \text{neg. sample}) - \text{sample response with inhibitor extract})}{(\text{Response of pos. control} - (\text{pos. sample} - \text{neg. sample}))} \times 100$$

The inhibition plots demonstrate that the allergens tested are inhibited by the relevant inhibitor extract in a concentration dependent fashion. Also, the target % inhibition of 50% for the highest inhibitor concentration tested was met. These results indicate specificity of *Aureobasidium pullulans*, *Blomia tropicalis*, Brazilian Peppertree, Budgerigar Feathers, Canary Feathers, Chicken Feathers, Loblolly Pine, nAsp r 1, nCan f 1, nDer f 1, nDer f 2, nDer p 1, nDer p 2, nFel d 1, nPen m 1, Parrot Feathers, *Penicillium brevicompactum*, Pigeon Droppings, Rabbit Bush, Tobacco, and Turkey Feather allergens.

Additional inhibition studies were conducted to show that the specific allergens are not cross-reacting to the unrelated allergens. Testing was performed using one positive sample with three unrelated allergen extracts at 1 mg/ml. A negative sample was used to measure the background response. Results on the positive sample for the following specific allergens were below 7% except for Loblolly Pine: *Aureobasidium pullulans*, *Blomia tropicalis*, Brazilian Peppertree, Budgerigar Feathers, Canary feathers, Chicken Feathers, nAsp r 1, nCan f 1, nDer f 1, nDer f 2, nDer p 1, nDer p 2, nFel d 1, nPen m 1, Parrot feathers, *Penicillium brevicompactum*, Pigeon Droppings, Rabbit bush, Tobacco, Turkey feathers:

Cross-reactivity: The manufacturer states there is no detectable crossreactivity with human serum immunoglobulins IgG, IgA, IgM or IgD at normal physiological levels.

- f. *Assay cut-off:*
Not applicable
2. Comparison studies:
 - a. *Method comparison with predicate device:*
Refer to Clinical studies
 - b. *Matrix comparison:*
Not applicable. Serum is the only matrix.
3. Clinical studies:
 - a. Clinical Sensitivity and specificity
Clinical performance of the IMMULITE® 2000 3gAllergy Specific IgE assay

for *Aureobasidium pullulans*, *Blomia tropicalis*, Brazilian Peppertree, Budgerigar Feathers, Canary feathers, Chicken Feathers, Loblolly Pine, nAsp r 1, nCan f 1, nDer f 1, nDer f 2, nDer p 1, nDer p 2, nFel d 1, nPen m 1, Parrot feathers, *Penicillium brevi-compactum*, Pigeon Droppings, Rabbit bush, Tobacco, Turkey feathers allergens was demonstrated by testing samples from non-atopic and atopic individuals. Atopic patients were selected from patients who had clinical documentation of allergy to specific allergen(s) or allergen group of interest and/or positive skin prick/ scratch test to specific allergen(s) of interest evaluated as 2+ or greater. Information on the skin test allergen extracts (crude or purified) was not documented. Non-atopic patients were clinically known non-allergenic or total IgE <130 ng/mL or 54 IU/mL (2.4 ng = 1 IU). Testing was performed on 140 samples for *Aureobasidium pullulans*, 154 samples for *Blomia tropicalis*, 150 samples for Brazilian Peppertree, 130 samples for Budgerigar Feathers, 131 samples for Canary feathers, 130 samples for Chicken Feathers, 150 samples for Loblolly Pine, 136 samples for nAsp r 1, 215 samples for nCan f 1, 170 samples for nDer f 1, 170 samples for nDer f 2, 168 samples for nDer p 1, 168 samples for nDer p 2, 161 samples for nFel d 1, 133 samples for nPen m 1, 131 samples for Parrot feathers, 138 samples for *Penicillium brevi-compactum*, 130 samples for Pigeon Droppings, 150 samples for Rabbit bush, 155 samples for Tobacco, and 130 samples for Turkey feathers.

Sensitivity and specificity of the new device, based on diagnosis of atopic status, are shown in the tables below:

<u>Allergen:</u> <i>Aureobasidium pullulans</i>		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	20	0	20
	negative	20	100	120
	Total	40	100	140

Sensitivity: 50% (20/40) (95% CI: 35-65%)

Specificity: 100 % (100/100)

<u>Allergen:</u> <i>Blomia tropicalis</i>		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	31	4	35
	negative	23	96	119
	Total	54	100	154

Sensitivity: 57% (31/54) (95% CI: 44-71%)

Specificity: 96 % (96/100) (95% CI: 92-100 %)

<u>Allergen:</u> Brazilian Peppertree		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	43	2	45
	negative	7	98	105

	Total	50	100	150
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Sensitivity: 86% (43/50) (95% CI: 76-96%)

Specificity: 98% (98/100) (95% CI: 95-101%)

<u>Allergen:</u> Budgerigar Feathers		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	14	0	14
	negative	16	100	116
	Total	30	100	130

Sensitivity: 47% (14/30) (95% CI: 29-65%)

Specificity: 100% (100/100)

<u>Allergen:</u> Canary feathers		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	21	4	25
	negative	9	97	106
	Total	30	101	131

Sensitivity: 70% (21/30) (95% CI: 54-86%)

Specificity: 96% (97/101) (95% CI: 92-100%)

<u>Allergen:</u> Chicken Feathers		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	18	1	19
	negative	12	99	111
	Total	30	100	130

Sensitivity: 60% (18/30) (95% CI: 42-78%)

Specificity: 99% (99/100) (95% CI: 97-100%)

<u>Allergen:</u> Loblolly Pine		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	34	0	34
	negative	16	100	116
	Total	50	100	150

Sensitivity: 68% (34/50) (95% CI: 55-81%)

Specificity: 100% (100/100)

<u>Allergen:</u> nAsp r 1		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	14	1	15
	negative	22	99	121
	Total	36	100	136

Sensitivity: 39% (14/36) (95% CI: 23-55%)

Specificity: 99% (99/100) (95% CI: 97-100%)

<u>Allergen:</u> nCan f 1		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	40	2	
	negative	1	172	
	Total	41	174	

Sensitivity: 98% (40/41) (95% CI: 93-102%)

Specificity: 99% (172/174) (95% CI: 97-100%)

<u>Allergen:</u> nDer f 1		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	47	4	51
	negative	10	109	119
	Total	57	113	170

Sensitivity: 82% (47/57) (95% CI: 72-92%)

Specificity: 96% (109/113) (95% CI: 93-100%)

<u>Allergen:</u> nDer f 2		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	46	4	50
	negative	11	109	120
	Total	57	113	170

Sensitivity: 81% (46/57) (95% CI: 70-91%)

Specificity: 96% (109/113) (95% CI: 93-100%)

<u>Allergen:</u> nDer p 1		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	49	3	52
	negative	7	109	116
	Total	56	112	168

Sensitivity: 88% (49/56) (95% CI: 79-96%)

Specificity: 97% (109/112) (95% CI: 94-100%)

<u>Allergen:</u> nDer p 2		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	51	2	53
	negative	5	110	115
	Total	56	112	168

Sensitivity: 91% (51/56) (95% CI: 84-99%)

Specificity: 98% (110/112) (95% CI: 96-101%)

<u>Allergen:</u> nFel d 1		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	48	5	53
	negative	7	101	108
	Total	55	106	161

Sensitivity: 87% (48/55) (95% CI: 78-96%)
 Specificity: 95% (101/106) (95% CI: 91-100%)

<u>Allergen:</u> nPen m 1		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	29	0	29
	negative	4	100	104
	Total	33	100	133

Sensitivity: 88% (14/31) (95% CI: 77-99%)
 Specificity: 100% (100/100)

<u>Allergen:</u> Parrot feathers		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	15	1	16
	negative	15	100	115
	Total	30	101	131

Sensitivity: 50% (15/30) (95% CI: 32-68%)
 Specificity: 99% (97/100) (95% CI: 97-100%)

<u>Allergen:</u> Penicillium brevi-compactum		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	23	0	23
	negative	15	100	115
	Total	38	100	138

Sensitivity: 61% (23/38) (95% CI: 45-76%)
 Specificity: 100% (100/100)

<u>Allergen:</u> Pigeon Droppings		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	16	0	16
	negative	14	100	114
	Total	30	100	130

Sensitivity: 53% (16/30) (95% CI: 35-71%)
 Specificity: 100% (100/100)

<u>Allergen:</u> Rabbit bush		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	38	4	42
	negative	12	96	108
	Total	50	100	150

Sensitivity: 76% (38/50) (95% CI: 64-88%)
 Specificity: 96% (96/100) (95% CI: 92-100%)

<u>Allergen:</u> Tobacco		Clinical Diagnosis		
		Atopic	Non-atopic	Total

IMMULITE 2000	positive	31	0	
	negative	24	100	
	Total	55	100	

Sensitivity: 56% (31/55) (95% CI: 43-69%)

Specificity: 100% (100/100)

Allergen: Turkey feathers		Clinical Diagnosis		
		Atopic	Non-atopic	Total
IMMULITE 2000	positive	20	1	21
	negative	10	99	109
	Total	30	100	130

Sensitivity: 67% (20/30) (95% CI: 50-84%)

Specificity: 99% (99/100) (95% CI: 97-101%)

Literature support was provided on allergens with low prevalence and % sensitivity as shown below:

Specific Allergen/ % Clinical Sen.	Allergen Name	Literature(s)	General Prevalence	Clinical Sensitivity
A3050 / 39%	nAsp r 1	1. Mari A, Schneider P, Wally V, Breitenbach M, Simon-Nobbe B. Sensitization to fungi: epidemiology, comparative skin tests, and IgE reactivity of fungal extracts. Clin Exp Allergy. 2003 Oct;33(10):1429-38. 2. Niedoszytko M, Chelminska , Jassem E, Czestochowska E. Association between sensitization to Aureobasidium pullulans (Pullularia sp) and severity of asthma. Ann Allergy Asthma Immunol. 2007 Feb;98(2):153-6	Fungi - 19.1% (621 out of 3248) within an atopic population ¹	Aspergillus 2.4-12.6% (78/3248 within allergic population) to (78/621 within fungi sensitive population) ¹ Aspergillus 20% (21/105 within Asthmatic population) ²
E78 / 47%	Budgerigar Feathers	1. Niedoszytko M, Chelminska , Jassem E, Czestochowska E. Association between sensitization to Aureobasidium pullulans (Pullularia sp) and severity of asthma. Ann Allergy Asthma Immunol. 2007 Feb;98(2):153-6 2. Kilpio K, Makinen-Kiljunen S, Haahtela T, Hannuksela M. Allergy to Feathers. Allergy 1998;53:159-164.	Feathers – 10-22% (3/30 control population - 22/105 within Asthmatic population) ¹	Hen, Duck, Goose Feathers – 28% (51/177 within atopic population) ²
M12 / 50%	Aureobasidium pullulans	1. Mari A, Schneider P, Wally V, Breitenbach M, Simon-Nobbe B. Sensitization to fungi: epidemiology, comparative skin tests, and IgE reactivity of fungal extracts. Clin Exp Allergy. 2003 Oct;33(10):1429-38. 2. Niedoszytko M, Chelminska , Jassem E, Czestochowska E. Association between sensitization to Aureobasidium pullulans (Pullularia sp) and severity of asthma. Ann Allergy Asthma Immunol. 2007 Feb;98(2):153-6	Fungi - 19.1% (621 out of 3248) within an atopic population ¹	Aureobasidium pullulans 27% (28/105 within Asthmatic population) ²

Specific Allergen/ % Clinical Sen.	Allergen Name	Literature(s)	General Prevalence	Clinical Sensitivity
E91 / 50%	Parrot Feathers	1. Niedozytko M, Chelminska , Jassem E, Czestochowska E. Association between sensitization to Aureobasidium pullulans (Pullularia sp) and severity of asthma. Ann Allergy Asthma Immunol. 2007 Feb;98(2):153-6 2. Kilpio K, Makinen-Kiljunen S, Haahtela T, Hannuksela M. Allergy to Feathers. Allergy 1998;53:159-164.	Feathers – 10 - 22% (3/30 control population - 23/105 within Asthmatic population) ¹	Hen, Duck, Goose Feathers – 28% (51/177 within atopic population) ²

b. Other clinical supportive data (when a. is not applicable):

Not applicable.

4. Clinical cut-off:

Not applicable.

5. Expected values/Reference range:

Not detected.

Refer to the Hoffman's 'Standard' and 'Extended Standard' classification system utilizing Class 0 to Class IV cut-offs (see Tables I and II below).

Table I: The Standard classification system utilizes the following class cutoffs:

Class	kU/L	Reactivity for Individual/Component Allergen(s)
0*	< 0.10	Absent or ND [†]
	0.10 – 0.34	Very Low
I	0.35 – 0.69	Low
II	0.70 – 3.49	Moderate
III	3.50 – 17.49	High
IV	17.5 – 52.49	Very High
V	52.5 – 99.99	
VI	≥ 100	

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

[†] ND: not detectable by IMMULITE 2000 3gAllergy.

Table II: The Extended standard classification system utilizes the following class cutoffs.

Class	kU/L	Reactivity for Individual/Component Allergen(s)
0	< 0.10	Absent or ND [†]
0/1	0.10 – 0.24	Very Low
I	0.25 – 0.39	Low
II	0.40 – 1.29	Moderate
III	1.30 – 3.89	High
IV	3.90–14.99	Very High
V	15.00– 24.99	
VI	≥ 25	

[†] ND: not detectable by IMMULITE 2000 3gAllergy

The choice of classification systems can be made by the user within the IMMULITE 2000 operational software.

Reference: Hoffman, DR. Comparison of methods of performing the Radioallergosorbent test: Phadebas, Fadal-Nalebuff and Hoffman protocols. Ann Allergy. 1980 Dec; 45(6)

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.