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

## I Background Information:

### A 510(k) Number

K260591

### B Applicant

Hycor Biomedical

### C Proprietary and Established Names

NOVEOS Specific IgE (sIgE), Capture Reagent F010, Sesame Seed (*Sesamum indicum*)
NOVEOS Specific IgE (sIgE), Capture Reagent F020, Almond (*Amygdalus communis*)
NOVEOS Specific IgE (sIgE), Capture Reagent F078, Bos d 8 Casein, Milk
NOVEOS Specific IgE (sIgE), Capture Reagent F080, Lobster (*Homarus gammarus*)
NOVEOS Specific IgE (sIgE), Capture Reagent F422, Ara h 1, Peanut

### 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:

Five new devices

### B Measurand:

Allergen specific IgE to Sesame Seed - F010, *Sesamum indicum*
Allergen specific IgE to Almond - F020, *Amygdalus communis*
Allergen specific IgE to Bos d 8 Casein, Milk - F078
Allergen specific IgE to Lobster - F080, *Homarus gammarus*
Allergen specific IgE to Ara h 1, Peanut - F422, Ara h 1

### C Type of Test:

Chemiluminescent, Quantitative

## III Intended Use/Indications for Use:

### A Intended Use(s):

See Indications for Use below.

### B Indication(s) for Use:

The NOVEOS Specific IgE assay is an *in vitro* quantitative assay for the measurement of allergen specific IgE in human serum. NOVEOS Specific IgE assay is to be used with the NOVEOS Immunoassay Analyzer. It is intended for use as an *in vitro* diagnostic aid in the clinical diagnosis of IgE mediated allergic disorders in conjunction with other clinical findings and is to be used in clinical laboratories.

### C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

### D Special Instrument Requirements:

For use on the NOVEOS Immunoassay Analyzer

## IV Device/System Characteristics:

### A Device Description:

The NOVEOS Specific IgE assay consists of the following reagents:

- IgE Common Kit:
  - Diluent A (human serum albumin in buffer with preservative)
  - Conjugate IgE (horse radish peroxidase labeled mouse monoclonal anti-IgE antibody with preservative)
  - Substrate A
  - Substrate B
  - Fluo Beads (streptavidin coated magnetic particles with preservative)

- Capture Reagent Pack
  - Sesame Seed - F010, *Sesamum indicum*
  - Almond - F020, *Amygdalus communis*
  - Bos d 8 Casein, Milk - F078
  - Lobster - F080, *Homarus gammarus*

- Ara h 1, Peanut - F422, Ara h 1

- IgE Calibrator Set (six levels: 0.07, 0.35, 0.70, 3.5, 17.5, 100 kU/L)
- IgE Calibrator Antibody Pack (mouse monoclonal anti-human IgE with preservative)
- IgE Positive Control Pack (known to be >0.35 kU/L)
- IgE Negative Control Packs (known to be <0.35 kU/L)
- Others:
  - Probe Wash Pack (phosphate buffered citric acid solution with preservative)
  - Wash Buffer Concentrate Pack (10X phosphate buffer saline solution with preservative)
  - Cuvette Wash Pack (citric acid solution with preservative)

**B Principle of Operation:**

The NOVEOS Specific IgE assay is an immunometric, chemiluminescent procedure for the quantitative determination of IgE of known specificity in human serum samples. It employs fluorescent labelled magnetic, streptavidin coated microparticles which are incubated with a biotinylated allergenic capture reagent, patient sample and monoclonal anti-human IgE antibody labeled with horseradish peroxidase (Conjugate IgE). The beads are collected by using a magnetic field to remove the liquid and then undergo a wash step to remove unbound biotinylated allergen prior to incubation with the sample. If present in the sample, sIgE binds to the captured biotinylated allergen. After incubation, the beads are washed and subsequently incubated with an horseradish peroxidase (HRP)-labeled anti-IgE monoclonal antibody (Conjugate IgE) to form an antibody-conjugate complex that is bound to the captured biotinylated allergen. After washing away the non-bound conjugate, a chemiluminescent substrate is added to the assay mixture resulting in light generation in proportion to the amount of antibody-conjugate that has been bound to the beads. The fluorescence is used to correct for any bead loss during the assay, and the (corrected) chemiluminescence is compared to a calibration curve to provide quantification of bound sIgE. The higher the value of chemiluminescent signal detected by the instrument, the higher the amount of sIgE detected in the sample tested. The concentration of allergen specific IgE is determined from a standard curve, which is traceable to the World Health Organization (WHO) reference reagent serum Immunoglobulin E (IgE) 11/234.

## V Substantial Equivalence Information:

**A Predicate Device Name(s):**

ImmunoCAP Specific IgE Assay

**B Predicate 510(k) Number(s):**

K051218

## C Comparison with Predicate(s):

| Device & Predicate Device(s): | K260591 (Candidate Device) | K051218 (Predicate Device) |
|---|---|---|
| Device Trade Name | NOVEOS Specific IgE (sIgE) | ImmunoCAP Specific IgE |
| **General Device Characteristic Similarities** | | |
| Intended Use/ Indications For Use | The NOVEOS Specific IgE assay is an *in vitro* quantitative assay for the measurement of allergen specific IgE in human serum. NOVEOS Specific IgE assay is to be used with the NOVEOS Immunoassay Analyzer. It is intended for use as an *in vitro* diagnostic aid in the clinical diagnosis of IgE mediated allergic disorders in conjunction with other clinical findings and is to be used in clinical laboratories. | ImmunoCAP Specific IgE is an *in vitro* quantitative assay for the measurement of allergen specific IgE in human serum or plasma (EDTA or Na-Heparin). ImmunoCAP Specific IgE is to be used with instruments Phadia 100, Phadia 250, Phadia 1000, Phadia 2500 and Phadia 5000. It is intended for *in vitro* diagnostic use as an aid in the clinical diagnosis of IgE mediated allergic disorders in conjunction with other clinical findings, and is to be used in clinical laboratories. |
| Assay Type | Quantitative | Same |
| Traceability | World Health Organization (WHO) reference reagent serum Immunoglobulin E (IgE) 11/234 | Same |
| Calibration Method | Heterologous interpolation based on Total IgE calibration curve | Same |
| Reaction Temperature | 37°C | Same |
| Limit of Quantification | 0.10 kU/L | Same |
| **General Device Characteristic Differences** | | |
| Solid Phase | Magnetic microparticles | Cellulose derivative |
| Assay Principle | Chemiluminescent assay | Fluoroenzyme-immunoassay |
| Instrument(s) | NOVEOS Immunoassay Analyzer | Phadia 100, Phadia 250/1000/2500/5000 |
| Specimen Type | Serum | Serum or plasma (EDTA, Na-Heparin) |
| Specimen Volume | 4 µL | 40 µL |
| Detection Antibody | HRP-conjugated mouse anti-human IgE monoclonal antibody | β-Galactosidase-anti-human IgE (mouse monoclonal antibody) |
| Calibrator levels | 6 levels: 0.07, 0.35, 0.7, 3.5, 17.5, 100 kU/L | 6 levels: 0, 0.35, 0.7, 3.5, 17.5, 100 kU/L |
| Time to First Result | 1 hour and 45 minutes | 1 hour and 45 minutes to 2 hours and 30 minutes depending on instrument model |

## VI Standards/Guidance Documents Referenced:

The following Clinical and Laboratory Standards Institute (CLSI) guidelines were used:

- CLSI I/LA20-A3: Performance Characteristics, Quality Assurance, and Clinical Utility of Immunological Assays for Human Immunoglobulin E Antibodies and Defined Allergen Specificities, Approved Guideline – Third Edition
- CLSI EP05-A3: Evaluation of Precision of Quantitative Measurement Procedures; Approved Guideline – Third Edition.
- CLSI EP06, 2nd ed.: Evaluation of Linearity of Quantitative Measurement, Approved Guideline – Second Edition
- CLSI EP07, 3rd ed.: Interference Testing in Clinical Chemistry; Approved Guideline –Third Edition
- CLSI EP17-A2: Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures; Approved Guideline – Second Edition
- CLSI EP25, 2nd ed: Evaluation of Stability of In Vitro Medical Laboratory Test Reagent Reagents– Second Edition
- CLSI EP28-A3c: Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory; Approved Guideline – Third Edition
- CLSI EP37, Supplemental Tables for Interference Testing in Clinical Chemistry – First Edition

## VII Performance Characteristics (if/when applicable):

### A Analytical Performance:

1. Precision/Reproducibility:

   a. Within-laboratory imprecision

   The within-laboratory precision study was conducted per the CLSI EP05-A3 for each of five NOVEOS Specific IgE assays. A panel of four to five human sera that included at least one negative sample were tested in duplicate per run, two runs per day for 20 days using one reagent lot on one to two NOVEOS Immunoassay Analyzers (for a total of 80 measurements per sample except for the highest sample of F020, and second-highest sample of F078). The standard deviation (SD) and %CV of the within-run, between-run, between-day, and total within-laboratory imprecision were calculated for each sample. The 20-day within-laboratory precision results for each NOVEOS Specific IgE assay are summarized in the following tables:

| NOVEOS Specific IgE, F010, Sesame Seed | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Sample | Mean (kU/L) | N | Within-Run | | Between-Run | | Between-Day | | Total | |
| | | | SD | %CV | SD | %CV | SD | %CV | SD | %CV |
| 1 | 0.11 | 80 | 0.01 | 11.1 | 0.01 | 8.2 | 0.00 | 3.0 | 0.02 | 14.1 |
| 2 | 0.27 | 80 | 0.01 | 4.8 | 0.01 | 5.1 | 0.00 | 0.0 | 0.02 | 7.0 |
| 3 | 7.92 | 80 | 0.24 | 3.0 | 0.08 | 1.0 | 0.16 | 2.1 | 0.30 | 3.8 |
| 4 | 90.74 | 80 | 8.55 | 9.4 | 2.54 | 2.8 | 1.87 | 2.1 | 9.11 | 10.0 |

| NOVEOS Specific IgE, F020, Almond | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Sample | Mean (kU/L) | N | Within-Run | | Between-Run | | Between-Day | | Total | |
| | | | SD | %CV | SD | %CV | SD | %CV | SD | %CV |
| 1 | 0.11 | 80 | 0.01 | 7.3 | 0.01 | 6.0 | 0.01 | 10.7 | 0.02 | 14.2 |
| 2 | 0.37 | 80 | 0.02 | 6.7 | 0.02 | 5.4 | 0.00 | 0.0 | 0.03 | 8.6 |
| 3 | 1.72 | 80 | 0.11 | 6.5 | 0.00 | 0.0 | 0.04 | 2.4 | 0.12 | 7.0 |
| 4 | 10.86 | 80 | 0.44 | 4.1 | 0.32 | 2.9 | 0.43 | 3.9 | 0.69 | 6.4 |
| 5 | 78.07 | 81* | 7.89 | 10.1 | 0.00 | 0.0 | 5.43 | 7.0 | 9.57 | 12.3 |

*Sample 5 had one outlier removed on Day 18 and an extra run with two replicates was added.

| NOVEOS Specific IgE, F078, Bos d 8 Casein, Milk | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Sample | Mean (kU/L) | N | Within-Run | | Between-Run | | Between-Day | | Total | |
| | | | SD | %CV | SD | %CV | SD | %CV | SD | %CV |
| 1 | 0.24 | 80 | 0.02 | 6.8 | 0.00 | 0.7 | 0.02 | 6.8 | 0.02 | 9.6 |
| 2 | 0.46 | 80 | 0.02 | 4.4 | 0.01 | 1.5 | 0.01 | 2.9 | 0.03 | 5.5 |
| 3 | 44.30 | 86* | 2.05 | 4.6 | 2.23 | 5.0 | 4.28 | 9.7 | 5.24 | 11.8 |
| 4 | 79.39 | 80 | 6.48 | 8.2 | 4.86 | 6.1 | 0.00 | 0.0 | 8.10 | 10.2 |

* Sample 3 had one additional run (two replicates) on Day 2, for the sample run on Day 15, two additional runs (four replicates) were performed with a new sample.

| NOVEOS Specific IgE, F080, Lobster | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Sample | Mean (kU/L) | N | Within-Run | | Between-Run | | Between-Day | | Total | |
| | | | SD | %CV | SD | %CV | SD | %CV | SD | %CV |
| 1 | 0.21 | 80 | 0.02 | 8.9 | 0.01 | 2.7 | 0.00 | 0.0 | 0.02 | 9.3 |
| 2 | 0.27 | 80 | 0.01 | 4.2 | 0.01 | 3.2 | 0.01 | 2.2 | 0.02 | 5.7 |
| 3 | 1.23 | 80 | 0.05 | 4.1 | 0.07 | 5.8 | 0.00 | 0.0 | 0.09 | 7.1 |
| 4 | 86.36 | 80 | 3.11 | 3.6 | 4.97 | 5.8 | 0.00 | 0.0 | 5.86 | 6.8 |

| NOVEOS Specific IgE, F422, Ara h 1, Peanut | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Sample | Mean (kU/L) | N | Within-Run | | Between-Run | | Between-Day | | Total | |
| | | | SD | %CV | SD | %CV | SD | %CV | SD | %CV |
| 1 | 0.13 | 80 | 0.02 | 12.22 | 0.00 | 0.00 | 0.00 | 0.00 | 0.02 | 12.22 |
| 2 | 0.35 | 80 | 0.02 | 5.63 | 0.01 | 1.75 | 0.01 | 2.95 | 0.02 | 6.59 |
| 3 | 5.79 | 80 | 0.24 | 4.07 | 0.28 | 4.80 | 0.62 | 10.70 | 0.72 | 12.46 |
| 4 | 88.90 | 80 | 4.43 | 4.99 | 5.59 | 6.30 | 6.59 | 7.40 | 9.71 | 10.92 |

b. Lot-to-lot imprecision

A panel of four to five serum samples were tested with three different lots of each NOVEOS Specific IgE assay. The samples were tested in replicates of five per run, one run per day, for five days, to generate a total of 75 replicates per sample for all three lots. The results are summarized in the following tables:

| NOVEOS Specific IgE, F010, Sesame Seed | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sample** | **Mean (kU/L)** | **N** | **Within-Run** | | **Between-Day** | | **Between-Lot** | | **Total** | |
| | | | **SD** | **%CV** | **SD** | **%CV** | **SD** | **%CV** | **SD** | **%CV** |
| 1 | 0.14 | 75 | 0.01 | 10.3 | 0.02 | 13.7 | 0.00 | 0.0 | 0.02 | 17.1 |
| 2 | 0.48 | 75 | 0.03 | 5.4 | 0.03 | 6.6 | 0.00 | 0.0 | 0.04 | 8.5 |
| 3 | 0.99 | 75 | 0.04 | 4.3 | 0.07 | 6.7 | 0.00 | 0.0 | 0.08 | 8.0 |
| 4 | 7.74 | 75 | 0.22 | 2.8 | 0.32 | 4.1 | 0.00 | 0.0 | 0.39 | 5.0 |
| 5 | 75.40 | 75 | 2.59 | 3.4 | 1.54 | 2.0 | 1.07 | 1.4 | 3.19 | 4.2 |

| NOVEOS Specific IgE, F020, Almond | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sample** | **Mean (kU/L)** | **N** | **Within-Run** | | **Between-Day** | | **Between-Lot** | | **Total** | |
| | | | **SD** | **%CV** | **SD** | **%CV** | **SD** | **%CV** | **SD** | **%CV** |
| 1 | 0.10 | 75 | 0.01 | 12.2 | 0.01 | 4.9 | 0.00 | 0.0 | 0.01 | 13.2 |
| 2 | 0.47 | 75 | 0.02 | 4.0 | 0.01 | 3.2 | 0.00 | 0.0 | 0.02 | 5.1 |
| 3 | 1.69 | 75 | 0.07 | 4.1 | 0.02 | 0.9 | 0.00 | 0.1 | 0.07 | 4.2 |
| 4 | 10.45 | 75 | 0.46 | 4.4 | 0.30 | 2.9 | 0.00 | 0.0 | 0.55 | 5.2 |
| 5 | 80.44 | 75 | 3.33 | 4.1 | 2.77 | 3.4 | 0.00 | 0.0 | 4.33 | 5.4 |

| NOVEOS Specific IgE, F078, Bos d 8 Casein, Milk | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sample** | **Mean (kU/L)** | **N** | **Within-Run** | | **Between-Day** | | **Between-Lot** | | **Total** | |
| | | | **SD** | **%CV** | **SD** | **%CV** | **SD** | **%CV** | **SD** | **%CV** |
| 1 | 0.20 | 75 | 0.02 | 7.7 | 0.01 | 5.0 | 0.00 | 0.0 | 0.02 | 9.2 |
| 2 | 0.47 | 75 | 0.03 | 5.9 | 0.01 | 3.2 | 0.01 | 2.0 | 0.03 | 7.0 |
| 3 | 1.78 | 75 | 0.07 | 3.8 | 0.05 | 3.1 | 0.00 | 0.0 | 0.09 | 4.9 |
| 4 | 83.43 | 75 | 4.05 | 4.9 | 4.10 | 4.9 | 0.00 | 0.0 | 5.76 | 6.9 |

| NOVEOS Specific IgE, F080, Lobster | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sample** | **Mean (kU/L)** | **N** | **Within-Run** | | **Between-Day** | | **Between-Lot** | | **Total** | |
| | | | **SD** | **%CV** | **SD** | **%CV** | **SD** | **%CV** | **SD** | **%CV** |
| 1 | 0.20 | 75 | 0.02 | 9.1 | 0.01 | 5.1 | 0.01 | 4.4 | 0.02 | 11.4 |
| 2 | 0.27 | 79* | 0.02 | 5.7 | 0.01 | 2.6 | 0.00 | 0.0 | 0.02 | 6.2 |
| 3 | 0.58 | 75 | 0.03 | 4.6 | 0.04 | 6.2 | 0.00 | 0.0 | 0.05 | 7.8 |
| 4 | 1.18 | 75 | 0.06 | 4.9 | 0.00 | 0.0 | 0.02 | 1.7 | 0.06 | 5.2 |
| 5 | 80.97 | 75 | 4.28 | 5.3 | 3.77 | 4.7 | 2.15 | 2.6 | 6.09 | 7.5 |

*Sample 2 had one outlier removed on Day 3 and an extra run with five replicates was added.

| NOVEOS Specific IgE, F422, Ara h 1, Peanut | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Sample | Mean (kU/L) | N | Within-Run | | Between-Day | | Between-Lot | | Total | |
| | | | SD | %CV | SD | %CV | SD | %CV | SD | %CV |
| 1 | 0.18 | 90* | 0.01 | 6.2 | 0.02 | 12.4 | 0.00 | 0.0 | 0.02 | 13.8 |
| 2 | 0.42 | 75 | 0.03 | 6.4 | 0.01 | 2.0 | 0.00 | 0.0 | 0.03 | 6.7 |
| 3 | 0.52 | 90* | 0.03 | 5.9 | 0.05 | 10.1 | 0.00 | 0.0 | 0.06 | 11.7 |
| 4 | 2.57 | 90* | 0.13 | 5.0 | 0.16 | 6.1 | 0.00 | 0.0 | 0.20 | 7.9 |
| 5 | 89.69 | 75 | 3.89 | 4.3 | 3.49 | 3.9 | 0.00 | 0.0 | 5.23 | 5.8 |

* Samples 1, 3, and 4 have 6 total days of testing data.

2. Linearity:

Linearity of each NOVEOS Specific IgE assay was evaluated in accordance with the CLSI guideline I/LA20-A3. Three sets of linearity dilution panels including six levels were prepared by using three positive human serum samples (low, mid, and high) diluted with one negative serum sample. Three dilution panels overlapped and created the full dilution panel. Testing was performed using one lot of each NOVEOS Specific IgE Assay in four replicates of each sample. The linearity data analysis was performed individually for each NOVEOS Specific IgE Assay in accordance with the CLSI EP06, 2nd Edition. For each sample level in the dilution panels, the mean value of the measured values, the predicted value and the %deviation from linearity were calculated. The results for each NOVEOS Specific IgE Assay are shown in the tables below:

The linear range and the claimed analytical mearing range for each NOVEOS Specific IgE are shown in the table below:

| NOVEOS Specific IgE | Tested Linear Range (kU/L) | Analytical Measuring Range (kU/L) | Regression Equation |
|---|---|---|---|
| F010, Sesame Seed | 0.06 – 103.06 | 0.10 – 100.0 | y = 0.92x – 0.01 |
| F020, Almond | 0.09 – 99.68 | 0.10 – 99.0 | y = 0.96x + 0.00 |
| F078, Bos d 8 Casein, Milk | 0.09 – 124.53 | 0.10 – 100.0 | y = 0.98x + 0.00 |
| F080, Lobster | 0.05 – 97.34 | 0.10 – 97.0 | y = 0.90x – 0.03 |
| F422, Ara h 1, Peanut | 0.05 – 96.87 | 0.10 – 96.0 | y = 1.02x – 0.01 |

3. Analytical Specificity/Interference:

a. Inhibition study:

Immunological specificity of each NOVEOS Specific IgE assay was verified through competitive inhibition study according to the CLSI guideline I/LA20-A3. Positive samples with specific IgE concentration used in the study include: 1.61 kU/L for F010, Sesame Seed; 0.97 kU/L for F020, Almond; 5.16 kU/L for F078, Bos d 8 Casein, Milk; 14.36 kU/L for F080, Lobster; and 8.85 kU/L for F422, Ara h 1, Peanut.

The allergen solution (inhibitor) was diluted 1:1 into the positive sample and subsequently serially diluted five times in two-fold increments. Each diluted sample was evaluated for dose-dependent inhibition in replicates of four using one lot of NOVEOS

sIgE allergens F010 (Sesame Seed), F020 (Almond), F078 (Bos d 8 Casein, Milk), F080 (Lobster), and F422 (Ara h 1, Peanut). Greater than 85% inhibition was achieved for F010 at 25 µg/mL (88% inhibition), F020 at 5.75 µg/mL (87% inhibition), and 83% inhibition was observed for F078 at 50 µg/mL and F080 at 3.125 µg/mL. For F422, 70% inhibition was achieved at 50 µg/mL which is near the beginning of the observed plateau in the dose-response curve.

For the NOVEOS Specific IgE Assays F010, F020, F078, F080, and F422 the related and unrelated allergens tested were as follows:

- For F010 (Sesame Seed), the related allergen F027 (Beef) and the unrelated allergens D071(Lepidoglyphus destructor), E072 (Mouse Urine) and M005 (Candida albicans) were tested as potential inhibitors at a final concentration of at least ten times the highest concentration of F010 at which ≥85% inhibition was achieved. All allergen inhibitors were spiked into the positive serum sample at a 1:1 dilution and evaluated with one lot of NOVEOS Specific IgE Assay, F010. The results of the single-dose inhibition studies using one related allergen (F027) and three unrelated allergens showed ≤15% inhibition at a test concentration of 1000 µg/mL (F027, D071, E072, and M005). The inhibition studies indicate that the NOVEOS Specific IgE Assay contains the immunologically relevant allergen.

- For F020, Almond, the related allergen F003 (Atlantic Cod) and the unrelated allergens G001 (Grass Pollen Sweet Vernal), M006 (Alternaria alternata), and W002 (Western Ragweed Pollen) were tested as potential inhibitors at a final concentration of at least ten times the highest concentration of F020 that achieved ≥85% inhibition. All allergen inhibitors were spiked into the positive serum sample at a 1:1 dilution and evaluated with one lot of NOVEOS Specific IgE Assay, F020. The results of the single-dose inhibition studies using one related allergen (F003) and three unrelated allergens showed ≤15% inhibition at test concentrations of 555 µg/mL (F003), 970 µg/mL (G001), 830 µg/mL (M006) and 1225 µg/mL (W0002). The inhibition studies indicate that the NOVEOS Specific IgE Assay contains the immunologically relevant allergen.

- For F078, Bos d 8 Casein, Milk, the related allergen F422 (Ara h 1, Peanut) and the unrelated allergens E221 (Can f 3 Dog Serum Albumin, Dog), M218 (Asp f 1, Aspergillus fumigatus), and T215 (Bet v 1 PR-10, Birch) were tested as potential inhibitors at a final concentration of at least ten times the highest concentration of F078 that achieved 83% inhibition. All allergen inhibitors were spiked into the positive serum sample at a 1:1 dilution and evaluated with one lot of NOVEOS Specific IgE Assay, F078. The results of the single-dose inhibition studies using one related allergen (F422) and three unrelated allergens showed ≤15% inhibition at test concentrations of 600 µg/mL (F422), 1970 µg/mL (E221), 660 µg/mL (M218) and 600 µg/mL (T215). The inhibition studies indicate that the NOVEOS Specific IgE Assay contains the immunologically relevant allergen.

- For F080, Lobster, the related allergen F013 (Peanut) and the unrelated allergens E003 (Horse Epithelia), M006 (Alternaria alternata), and T218 (Virginia Live Oak Pollen) were tested as potential inhibitors at a final concentration of at least ten times

the highest concentration of F080 that achieved 83% inhibition. All allergen inhibitors were spiked into the positive serum sample at a 1:1 dilution and evaluated with one lot of NOVEOS Specific IgE Assay, F080. The results of the single-dose inhibition studies using one related allergen (F013) and three unrelated allergens showed ≤15% inhibition at test concentrations of 1745 µg/mL (F013), 555 µg/mL (E003), 830 µg/mL (M006) and 1325 µg/mL (T218). The inhibition studies indicate that the NOVEOS Specific IgE Assay, F080 contains the immunologically relevant allergen.

- For F422, Ara h 1, Peanut, the related allergen F077 (Bos d 5, β-lactoglobulin, Milk) and the unrelated allergens E221 (Can f 3, Dog Serum Albumin, Dog), M218 (Asp f 1, *A. fumigatus*), and T215 (Bet v 1 PR-10, Birch) were tested as potential inhibitors at a final concentration of at least ten times the concentration of F422 at which 70% inhibition was achieved. All allergen inhibitors were spiked into the positive serum sample at a 1:1 dilution and evaluated with one lot of NOVEOS Specific IgE Assay, F422. The results of the single-dose inhibition studies using one related allergen (F077, Bos d 5, β-lactoglobulin, Milk) and three unrelated allergens showed ≤15% inhibition at test concentrations of 3520 µg/mL (F077), 1970 µg/mL (E221), 1200 µg/mL (T215) and 660 µg/mL (M218). The inhibition studies indicate that the NOVEOS Specific IgE Assay contains the immunologically relevant allergen.

b. Interference:

*i.* Endogenous Substance Interference:

The effect of the presence of elevated levels of human serum albumin, hemoglobin, triglycerides, conjugated bilirubin, and unconjugated bilirubin in serum samples was evaluated by testing three serum sample pools (one negative above LoQ, one near the cut-off, and one positive) spiked with varying levels of each interferent. Testing was conducted with seven replicates in one assay run using one lot each of the NOVEOS sIgE assays for F010 (Sesame Seed), F020 (Almond), F078 (Bos d 8 Casein, Milk), F080 (Lobster), and F422 (Ara h 1, Peanut). The % Interference for each sample spiked with the potential interfering substance was calculated by comparing its result to that of the corresponding control sample spiked with an equal volume of the solvent without the interfering substance. No interference was noted for samples containing human serum albumin up to 121 g/L, hemoglobin up to 200 mg/dL, intralipid up to 3000 mg/dL, conjugated bilirubin up to 30 mg/dL, and unconjugated bilirubin up to 20 mg/dL. Interference testing with rheumatoid factor (RF) was previously performed for NOVEOS sIgE allergens and found no significant interference at 513 IU/mL (refer to K200825).

*ii.* Exogenous Substance Interference:

Refer to K200825.

c. Cross-reactivity:

The potential cross-reactivity of non-IgE (IgA, IgD, IgG, and IgM) to the NOVEOS sIgE assays was evaluated in K200825.

4. Assay Reportable Range:

The assay reportable range is the same as the claimed analytical measuring range for each NOVEOS sIgE as shown in the table below:

| NOVEOS Specific IgE | Assay Reportable Range/ Analytical Measuring Range |
|---|---|
| F010, Sesame Seed | 0.10 – 100.0 kU/L |
| F020, Almond | 0.10 – 99.0 kU/L |
| F078, Bos d 8 Casein, Milk | 0.10 – 100.0 kU/L |
| F080, Lobster | 0.10 – 97.0 kU/L |
| F422, Ara h 1, Peanut | 0.10 – 96.0 kU/L |

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

a. Traceability:

The IgE calibrators are traceable to the World Health Organization (WHO) third International Standard 11/234 of Human Serum Immunoglobulin E.

b. Kit stability:

The stability study was conducted in accordance with CLSI EP25-A2.

*Shelf-life stability:* A real-time stability study for the NOVEOS Specific sIgE Capture Reagent Pack of F010 (Sesame Seed), F020 (Almond), F078 (Bos d 8 Casein, Milk), F080 (Lobster), and F422 (Ara h 1, Peanut) stored at 2–8°C is ongoing using three lots. The test was planned by testing two positive samples and one negative sample. In addition, an accelerated stability study was conducted with two positive samples and one negative sample using three lots of each Capture Reagent stored at 37°C to predict a shelf-life stability of 36 months if stored as 2–8°C. All other reagents required to perform specific IgE testing on the NOVEOS system are not allergen specific and the shelf-life stability of these reagents has been previously established per K182479 and K191510 (for the Fluo Beads reagent).

*On-board stability:* A study was performed on one instrument using three samples (two positive and one negative or low-level sample) using at least one lot of each NOVEOS Specific IgE Capture Reagent Pack for F010 (Sesame Seed), F020 (Almond), F078 (Bos d 8 Casein, Milk), F080 (Lobster), and F422 (Ara h 1, Peanut). The testing was performed for each sample in three replicates at Day 0 and scheduled timepoints of 15, 28, and 29 days. The result supports that the allergen specific capture reagent is stable for 28 days stored on-board. The result also supports that the allergen specific Capture Reagent Packs once opened and stored at 2-8°C are stable up to 28 days. The on-board stability for all the other assay components has been previously established per K182479.

*Open-vial stability:* Both an ongoing real-time stability study and an accelerated stability studies were performed for the NOVEOS sIgE allergens F010 (Sesame Seed), F020

(Almond), F078 (Bos d 8 Casein, Milk), F080 (Lobster), and F422 (Ara h 1, Peanut). The studies evaluated opened capture reagent vials filled to 100% and 50% fill volume, stored at 37°C for up to 10 days to simulate the stress of real-time aging. The result supports that the allergen specific Capture Reagent Packs are stable for 15 days once opened and stored at 2-8°C.

6. Detection Limit:

The limit of blank (LoB), limit of detection (LoD), and limit of quantitation (LoQ) of each NOVEOS sIgE assay, i.e., F010, F020, F078, F080, and F422 on the NOVEOS Immunoassay Analyzer were determined according to CLSI EP17-A2.

The LoB was determined by testing four analyte-free human serum samples in replicates of five per run, over three runs, with no more than one run per day, using two lots of each capture reagent on one NOVEOS Immunoassay Analyzer. The LoB was estimated as the 95$^{th}$ percentile of the 60 measurements for each of the two lots tested.

The LoD was determined by testing protocol summarized in the table below and calculated based on CLSI EP17-A2:

| NOVEOS sIgE | No. of Samples | No. of Lots | No. of Analyzers | Testing Protocol |
|---|---|---|---|---|
| F010 | 5 | 2 | 1 | 5 samples x 5 replicates/run x 3 runs = 75/Lot* |
| F020 | 6 | 2 | 1 | 6 samples x 5 replicates/run x 3 runs = 90/Lot |
| F078 | 4 | 2 | 1 | 4 samples x 5 replicates/run x 3 runs = 60/Lot |
| F080 | 4 | 2 | 1 | Lot 1: 4 samples x 5 replicates/run x 3 runs = 61/Lot**<br>Lot 2: 4 samples x 5 replicates/run x 3 runs = 60/Lot** |
| F422 | 4 | 2 | 1 | 4 samples x 5 replicates/run x 3 runs = 60/Lot |

* One outlier was excluded from the dataset.

** A testing error resulted in missing replicates for Lot 1; an additional test run on additional day was added for both lots to ensure a minimum of 60 total replicates were run.

The LoQ was determined by testing four to six low IgE samples with five replicates per run over three testing days using two lots of each capture reagent on one NOVEOS Immunoassay Analyzer resulting in a total of 60 to 90 replicates across all samples per lot. Data analysis was performed according to the precision profile method using within-lab precision results. The LoQ is defined as the mean value of the lowest sample which fulfills the specification for the total within-laboratory imprecision (<20%CV).

The claimed LoB, LoD, and LoQ are based on the highest value obtained from the two lots tested and are summarized in the table below:

| NOVEOS Specific IgE | LoB (kU/L) | LoD (kU/L) | LoQ (kU/L) |
|---|---|---|---|
| F010, Sesame Seed | 0.08 | 0.09 | 0.10 |
| F020, Almond | 0.07 | 0.08 | 0.10 |
| F078, Bos d 8 Casein, Milk | 0.07 | 0.09 | 0.10 |
| F080, Lobster | 0.07 | 0.08 | 0.10 |
| F422, Ara h 1, Peanut | 0.07 | 0.08 | 0.10 |

7. Assay Cut-Off:

See clinical cut-off

## B Comparison Studies:

1. Method Comparison with Predicate Device:

Refer to clinical study section below

2. Matrix Comparison:

Not Applicable

## C Clinical Studies:

1. Clinical Sensitivity and Specificity:

For each NOVEOS Specific IgE assay, the clinical performance was evaluated by comparing the test results from a cohort of atopic and non-atopic samples to the clinical diagnosis of allergy. The atopic samples were from different U.S. and EU vendors with a clinical history of allergy-like symptoms and identified as positive by oral food challenge (OFC) or diagnosed positive by a physician. The non-atopic samples were from donors with no reported allergy and were deemed negative by ImmunoCAP testing (<0.35 kU/L). The sIgE test results are expressed as positive or negative using a cut-off of 0.35 kU/L. The clinical sensitivity and clinical specificity for each NOVEOS Specific IgE Assay are summarized in the following tables:

| | | Clinical Diagnosis | | |
|---|---|---|---|---|
| | | **Atopic** | **Non-atopic** | **Total** |
| **NOVEOS Specific IgE, F010, Sesame Seed** | **Positive** | 43 | 0 | 43 |
| | **Negative** | 5 | 121 | 126 |
| | **Total** | 48 | 121 | 169 |
| Clinical Sensitivity: 89.6% (43/48) (95% CI: 77.8 – 95.5%)<br>Clinical Specificity: 100.0% (121/121) (95% CI: 96.9 – 100.0%) | | | | |

| | | Clinical Diagnosis | | |
|---|---|---|---|---|
| | | **Atopic** | **Non-atopic** | **Total** |
| **NOVEOS Specific IgE, F020, Almond** | **Positive** | 40 | 0 | 40 |
| | **Negative** | 7 | 122 | 129 |
| | **Total** | 47 | 122 | 169 |
| Clinical Sensitivity: 85.1% (40/47) (95% CI: 72.3 – 92.6%)<br>Clinical Specificity: 100.0% (122/122) (95% CI: 96.9 – 100.0%) | | | | |

| | | Clinical Diagnosis | | |
|---|---|---|---|---|
| | | **Atopic** | **Non-atopic** | **Total** |
| **NOVEOS Specific IgE, F078, Bos d 8 Casein, Milk** | **Positive** | 24 | 0 | 24 |
| | **Negative** | 5 | 130 | 135 |
| | **Total** | 29 | 130 | 159 |
| Clinical Sensitivity: 82.8% (24/29) (95% CI: 65.5 – 92.4%)<br>Clinical Specificity: 100.0% (130/130) (95% CI: 97.1 – 100.0%) | | | | |

| | | Clinical Diagnosis | | |
|---|---|---|---|---|
| | | **Atopic** | **Non-atopic** | **Total** |
| **NOVEOS Specific IgE, F080, Lobster** | **Positive** | 41 | 0 | 41 |
| | **Negative** | 8 | 124 | 132 |
| | **Total** | 49 | 124 | 173 |
| Clinical Sensitivity: 83.7% (41/49) (95% CI: 71.0 – 91.5%)<br>Clinical Specificity: 100.0% (124/124) (95% CI: 97.0 – 100.0%) | | | | |

| | | Clinical Diagnosis | | |
|---|---|---|---|---|
| | | **Atopic** | **Non-atopic** | **Total** |
| **NOVEOS Specific IgE, F422, Ara h 1, Peanut** | **Positive** | 32 | 7 | 39 |
| | **Negative** | 22 | 139 | 161 |
| | **Total** | 54 | 146 | 200 |
| Clinical Sensitivity: 59.3% (32/54) (95% CI: 46.0 – 71.3%)<br>Clinical Specificity: 95.2% (139/146) (95% CI: 90.4 – 97.7%) | | | | |

The following literature that supports the observed sensitivity of the NOVEOS sIgE assay F422 Ara h 1, Peanut allergen is presented in the table below.

| **NOVEOS sIgE assay** | **Literature Citation(s)** | **Prevalence** | **Reported Performance** |
|---|---|---|---|
| F422, Ara h 1, Peanut<br>Sensitivity: 59.3%<br>Specificity: 95.2% | Gupta RS, et al. JAMA Network Open. 2019;2(1):e185630 | ~1.8% among US adults have reported peanut allergy | not reported |
| | Asarnoj A. et al. Allergy 2010;65: 1189-1195 | Among children sensitized to peanut, only 50% had IgE reactivity to Ara h 1. | |

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

   Not Applicable

**D 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 <0.70 | Positive with increasing sIgE concentration |
| II | 0.70 to <3.50 | |
| III | 3.50 to <17.50 | |
| IV | 17.50 to <50.00 | |
| V | 50.00 to <100.00 | |
| VI | ≥100 | |

**E Expected Values/Reference Range:**

The expected value is <0.35 kU/L (negative) for a specific allergen in an apparently healthy (non-atopic) individual. Each laboratory should establish its own expected value/reference range.

The reference range of NOVEOS Specific IgE assay for F010 (Sesame Seed), F020 (Almond), F078 (Bos d 8 Casein, Milk), F080 (Lobster), and F422 (Ara h 1, Peanut) in the normal population was evaluated based on the data collected in clinical studies which tested samples from apparently healthy subjects including: 121 non-atopic samples for F010; 122 non-atopic samples for F020; 130 non-atopic samples for F078; 124 non-atopic samples for F080; and 173 non-atopic samples for F422. All samples tested with the NOVEOS Specific IgE assays were below 0.35 kU/L.

## VIII Proposed Labeling:

The labeling does not support 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.