

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

**A. 510(k) Number:**

k081615

**B. Purpose for Submission:**

New device

**C. Measurand:**

Carcinembryonic antigen (CEA)

**D. Type of Test:**

Quantitative, paramagnetic particle (Dynabeads®) chemiluminescent immunoassay

**E. Applicant:**

Olympus America, Inc.

**F. Proprietary and Established Names:**

Olympus CEA assay

**G. Regulatory Information:**

1. Regulation section:

21 CFR 866.6010 Tumor-associated antigen immunological test system

21 CFR 862.1660 Quality Control material (assayed and unassayed)

21 CFR 862.1150 Calibrator

2. Classification:

Class II

3. Product code:

DHX, System, Test, Carcinoembryonic Antigen (CEA)

JJX, Single (Specified) Analyte Controls (Assayed and Unassayed)

JIT, Calibrator, Secondary

4. Panel:

Immunology (82)

Clinical Chemistry (75)

**H. Intended Use:**

1. Intended use(s):

The Olympus CEA assay is a paramagnetic particle (Dynabeads®), chemiluminescent immunoassay for the quantitative determination of carcinoembryonic antigen levels in human serum and lithium heparin plasma using the Olympus AU3000i Immunoassay System. The Olympus CEA assay is indicated for serial measurement of CEA as an aid in the management (monitoring) of colorectal cancer patients. These CEA values must be interpreted in conjunction with all other clinical and laboratory data before a medical decision is made. For *in vitro* diagnostic use only.

2. Indication(s) for use:

Same as Intended Use

3. Special conditions for use statement(s):

Prescription use only

4. Special instrument requirements:

Olympus AU3000i Immunoassay System

**I. Device Description:**

The device is an *in vitro* diagnostic device using a paramagnetic particle (Dynabeads®), chemiluminescent immunoassay principle for the quantitative measurement of CEA on Olympus AU3000i Immunoassay System instruments using human serum or lithium heparin plasma specimens. Each Olympus CEA kit contains reagents sufficient for 200 tests. The kit is comprised of two reagents, one calibrator, one control and one septum and one package insert as follows:

- R1: Paramagnetic particles coated with murine monoclonal anti-CEA antibody, 4, 5 Tris buffer, pH 7.3 with protein stabilizers, detergent and preservative.
- R2: Alkaline phosphatase labeled murine monoclonal anti-CEA antibody, 4, 5 MES buffer, pH 6.5 with protein stabilizers, detergent and preservative.
- Calibrator: CEA prepared in a bovine matrix with preservative.
- Control: CEA  $\approx 5 \mu\text{g/L}$  ( $\approx 5 \text{ ng/mL}$ ) prepared in human matrix with preservative.
- Septum

**J. Substantial Equivalence Information:**

1. Predicate device name(s):  
Roche Elecsys CEA Assay, Elecsys PreciControl Tumor Marker Control, Elecsys CEA CalSet.
2. Predicate 510(k) number(s):  
k964368/k980887/k050387/k964368
3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Intended Use	For the quantitative determination of carcinoembryonic antigen levels in human serum or lithium heparin plasma	Same
Traceability/Standardization	First IRP WHO Reference Standard 73/601	Same
Measurement	Quantitative	Same
Assay Similarities	Chemiluminescence, Sandwich principle	Same
Analyte	CEA	CEA
Antibody R2	Mouse monoclonal anti-CEA	Same
Solid Phase	Microparticle	Same
Reagent Storage Form	Liquid	Same
Control Matrix	Prepared in human matrix	Same
Calibrator Constituent	Single	Same
Control and Calibrator Storage form	Liquid	Same
<b>Method</b>	Automated	Same

Differences
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Item	Device	Predicate
Instrument Required	Olympus AU3000i Immunoassay System	Roche Elecsys 2010
Assay Technology	paramagnetic particle (Dynabeads®), chemiluminescent immunoassay	Electrochemiluminescence (ECLIA)
Indications for use	For serial measurement of CEA as an aid in the management (monitoring) of colorectal cancer patients	For serial measurement of CEA to aid in the management of cancer patients
Specimen	Serum or Lithium heparin plasma	Serum or sodium heparin or potassium EDTA or sodium citrate plasma
Antibody R1	Mouse monoclonal anti-CEA	Biotinylated anti-CEA antibody (mouse/human)
Measurement range	0.05 to 500.00 ng/mL	0.200 to 1000 ng/ml
Control and Calibrator Levels	One	Two
Sample Volume	50 µL	10 µL
Limit of detection	0.08 ng/ml	0.20ng/ml
Control Stability	2-8°C for 28 days	2-8°C for 14 days
Calibrator Stability	2-8°C for 28 days	2-8°C for 12 weeks
On board reagent stability	28 days	6 weeks (Elecsys 2010)
Calibrator Matrix	Bovine matrix	Buffer/protein matrix
Solid Phase binding principle	Direct Coating	Biotin and Streptavidin

**K. Standard/Guidance Document referenced (if applicable):**

CLSI EP5-A2; Evaluation of Precision Performance of quantitative measurement methods; Approved Guideline – Second Edition.  
 CLSI EP9 – A2, Method Comparison and Bias Estimation Using Patient Samples  
 CLSI EP7-A2; Interference testing in clinical chemistry  
 CLSI C28-A2 How to Define and Determine Reference Intervals in the Clinical Laboratory.  
 CLSI EP17-A. Protocols for Determination of Limits of Detection and Limits of Quantitation.

**L. Test Principle:**

The Olympus CEA assay is a two-step paramagnetic particle enzyme immunoassay. It is based on the sandwich assay principle and used to quantitative CEA in human serum and lithium heparin plasma. The Olympus CEA assay reagent and sample are added to the assay cuvette in the following sequence:

1. Samples are incubated with a monoclonal anti-CEA antibody bound to paramagnetic particles.
2. **After a washing step**, a second monoclonal anti-CEA antibody conjugated with alkaline phosphatase is added. The CEA reacts with the paramagnetic particles and

the conjugated antibody to form a sandwich complex. The **washing steps** remove the unbound material.

3. The chemiluminescent substrate is added to the assay cuvette and reacts with the bound alkaline phosphatase (ALP). Light generated by the reaction is measured by the luminometer. The light emission is proportional to the quantity of CEA in the sample.

4. Results are calculated from a predefined calibration curve. The Olympus AU3000i system automatically calculates the CEA concentration of each sample in µg/L or ng/mL.

**M. Performance Characteristics (if/when applicable):**

1. Analytical performance:

*a. Precision/Reproducibility:*

The expected working imprecision of the Olympus CEA assay is designed to be ≤5% (Within Laboratory CV). Precision was determined at 4 laboratories using multiple lots, and 7 levels of pooled human sera and/or CEA controls according to CLSI protocol EP5-A2: 2 runs in duplicate per day for 20 days (n = 80) except where noted.

Pools	Site	Mean [µg/L],[ng/mL]	Repeatability (Within Run)		Within Laboratory (Total)	
			SD [µg/L],[ng/mL]	CV (%)	SD [µg/L],[ng/mL]	CV (%)
1	1	1.674	0.026	1.6	0.042	2.5
	2	1.676	0.029	1.7	0.053	3.1
	3	1.667	0.024	1.4	0.045	2.7
	4	1.745	0.033	1.9	0.070	4.0
2	1	25.952	0.396	1.5	0.600	2.3
	2	26.021	0.263	1.0	0.661	2.5
	3	25.586	0.344	1.3	0.760	3.0
	4	25.839	0.465	1.8	0.883	3.4
3	1	193.733	2.965	1.5	5.874	3.0
	2	201.067	4.278	2.1	7.648	3.8
	3	189.745	4.042	2.1	8.411	4.4
	4	190.942	4.757	2.5	8.747	4.6
4*	1	475.348	12.9	2.7	20.9	4.4
Control Level 1	4	3.333	0.061	1.8	0.107	3.2
Control Level 2	4	44.163	0.823	1.9	1.607	3.6
Test Kit Control	3	4.632	0.069	1.5	0.104	2.2

\* Note: The additional 4th level sample was tested twice per day for 5 days.

Lot-to-Lot Reproducibility.

Protocol: Three serum samples [High (210.0 µg/L or ng/mL), Medium (26.0 µg/L or ng/mL) and Low (1.8 µg/L or ng/mL)] were tested in parallel in two different lots. The difference in value divided by the lowest recovery value was calculated for each sample.

Acceptance criterion = 10%.

Results: results for all three samples were less than 10%. Thus between lot reproducibility met manufacturer specifications.

*b. Linearity/assay reportable range:*

Linearity: The testing was conducted using human serum samples with a base level of CEA equal to 3.99 micrograms/L. The samples were further spiked with antigen and then diluted using sample diluent SDIL 1. Concentrations tested were 3.16, 256.57, 355.15, and 527.80 ng/mL as well as numerous samples around zero. Acceptable deviation was set at 1.2 ng/mL or 30% according to the Wisconsin State Laboratory of Hygiene (WSLH). The graph presented on page 226 of the original submission shows good linearity up to upper end of the declared measurement range determined by the high calibrator (500 ng/L).

Dilution: To demonstrate dilution linearity of the assay, three patient samples were diluted to 4 levels with a prepared standard solution built with a bovine serum matrix. A neat (undiluted) sample was also run. Percent recovery is calculated by comparing the observed CEA result with the expected value. Recoveries within 10% of the expected results for the overall/total mean recovery for a given sample is considered acceptable. .

Results:

Sample	Dilution	Expected [µg/L]/[ng/mL]	Observed [µg/L]/[ng/mL]	Recovery [%]
1	-	-	26.37	-
	1:2	13.19	13.32	101.00
	1:4	6.59	6.87	104.24
	1:8	3.30	3.54	107.52
	1:16	1.65	1.75	106.13
	Mean	-	-	104.72
2	-	-	100.82	-
	1:2	50.41	50.51	100.20
	1:4	25.21	25.71	102.01
	1:8	12.60	13.32	105.72
	1:16	6.30	7.00	111.06
	Mean	-	-	104.75
3	-	-	252.87	-
	1:2	126.44	122.53	96.91
	1:4	63.22	60.24	95.29
	1:8	31.61	30.01	94.95
	1:16	15.80	15.43	97.66
	Mean	-	-	96.20

Conclusion: The recovery ranges were 94.95% to 111.06% with a mean of 101.89% for three diluted human serum samples with original concentrations between 26.37 and 252.87 µg/L (26.37 and 252.87 ng/mL) of CEA. Thus the assay demonstrates acceptable dilution recovery in the lower half of their declared measurement range.

Spiked Recovery Studies: Three human serum pools with endogenous analyte levels of 2.65 to 4.22 µg/L (2.65 to 4.22 ng/mL) were spiked with three different known levels of CEA. The actual percentage of antigen recovered is compared to the theoretical amount spiked into the samples. Recoveries within 10% of the expected overall mean recovery were considered acceptable. The recoveries ranged from 85.32% to 101.35% with a mean of 93.24%. One sample pool appeared to have an assay interferent of some kind, while two specimen pools were acceptable.

Sample	Amount added [µg/L],[ng/mL]	Observed [µg/L],[ng/mL]	Recovery [%]
1	-	2.65	-
	9.99	10.94	86.53
	49.93	44.87	85.32
	494.52	469.59	94.45
	Mean	-	88.77

2	-	2.79	-
	9.99	12.25	95.89
	49.93	50.46	95.70
	494.52	504.03	101.35
	Mean	-	97.65

3	-	4.22	-
	9.99	12.89	90.70
	49.93	49.21	90.88
	494.52	490.60	98.37
	Mean	-	93.31

Samples can be accurately measured within the measuring range of the LOQ (0.08 ng/mL) and the highest calibrator value (500 ng/mL)

The linearity/recovery data demonstrates that the test gives acceptable accuracy.

Hook Effect: A concentrated sample of purified CEA antigen was measured both neat and on dilution within the measuring range of the CEA assay. There was no high dose effect observed at CEA concentrations up to 375,000 µg/L (375,000 ng/mL). This assay uses a two-step design that significantly reduces the risk of a high dose hook effect.

- c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*  
Traceability. Assay calibrators are traceable to the First IRP WHO Reference

Standard 73/601 for human CEA.

Product Stability on Board: Reagent bottles from two lots were placed on board the instrument and tested periodically over a 28-day period for calibrator and control recovery. Serum samples across the measuring range (QC Panel) were tested at the end of the time period to compare on-board reagents with fresh reagents. The % drift from Day 0 at each time point was calculated from calibrator and control recoveries. The % differences between the fresh and on-board QC Panel recoveries were calculated. The percent drift did not exceed the acceptance criterion of 10%. The assay can sit on board the analyzer for 28 days

Calibration Stability: Reagent bottles from two lots were placed on board the instrument and tested periodically during the on-board study for calibrator and control recovery. The % drift from Day 0 at each time point was calculated from calibrator and control recoveries. The percent drift did not exceed the acceptance criterion of 10%. The calibrators can sit on board the analyzer for 28 days. The instrument uses the calibrator to define the calibration curve.

Calibrator and Control Open Vial Stability: Vials of calibrator and control were opened on day 0 and tested periodically during the on-board study. The % drift from Day 0 at each time point was calculated from calibrator and control recoveries. The percent drift did not exceed the acceptance criterion of 10%. The calibrators and controls can sit open on board the analyzer for 28 days. The control target values are encoded in the bar codes and accessible through the system

Real Time Product Stability. Kits from three lots were stored real-time according to the directions for use at 2 to 8°C. The stability was tested with quality control samples across the measurable range at time 0, 6, 7 and 12 months. . Consistent performance (within 10% drift) was demonstrated during the tested shelf life period. The data so far demonstrates a 12 month stability period. These studies are ongoing to extend the shelf-life period, if possible.

*d. Detection limit:*

The limits of blank, detection, and quantitation of CEA on the Olympus AU3000i were determined according to CLSI protocol EP17-A.

Limit of Blank (LOB)

The limit of blank was obtained by running 60 replicates of the blank sample. The limit of blank is the 95<sup>th</sup> percentile of the blank samples = 0.0000 µg/L.

The Limit of Detection (LOD)

The LOD, i.e., the lowest amount of analyte that can be detected with 95% probability, for CEA is based on the levels of 5 serum samples which were tested in duplicate . The LOD was determined to be 0.0038 µg/L and far exceeded expectations of 0.05 µg/L.

Limit of Quantitation

The Limit of Quantitation (LOQ) for the Olympus CEA assay was determined to be 0.08 µg/L (0.08 ng/mL). This was determined according to CLSI protocol EP17-A and represents the lowest concentration of CEA that can be measured with a total imprecision of 19.5%.

*e. Analytical specificity:*

The following cross-reactivities were determined for the assay by adding a predefined amount of potential cross-reactants to a human sample.

<b>Cross-reactant</b>	<b>Concentration Tested [µg/L],[ng/mL]</b>	<b>% Cross-reactivity</b>
Non-specific Cross-reacting Antigen 1 (NCA1)	200	0.42
Non-specific Cross-reacting Antigen 2 (NCA2)	20	24.81
AFP	1000	0.82

Cross-reactivity to NCA2 was evident.

### Interferences

Summary of Studies Performed. The interferents assessed were Bilirubin (Icteric), Haemolysate, Intralipid™ (Lipemis), Human anti-mouse antibody (HAMA) and Rheumatoid factor (RF). The Bilirubin (Icteric), Haemolysate and Lipemia interference studies were carried out by adding increasing amounts of each interferent to a human serum pool containing CEA. These samples, and a control sample containing no added interferents, were measured using the Olympus CEA assay. The HAMA and RF interference studies were carried out by measuring commercially available samples on the Olympus AU3000i™ Immunoassay System and on a reference system, Roche Elecsys. One lot of reagent was tested.

Results of studies conducted to evaluate the susceptibility of the method to interference were as follows:

Icterus: Interference less than 3% up to 40 mg/dL or 684 µmol/L bilirubin (unconjugated)

Lipemia: Interference less than 5% up to 10 g/L Intralipid®\*

Hemolysis: Interference less than 7% up to 5 g/L hemoglobin

Rheumatoid factor: No significant interference up to 2010 IU/mL of rheumatoid factor.

HAMA: No significant interference in two samples known to contain HAMA.

\*Intralipid is a 20% IV fat emulsion used to emulate extremely turbid samples. Approximate triglyceride concentration is 30 g/L.

The following limitation statement was added to the package insert to mitigate the risk of interference from endogenous interferences: “As with all tests containing monoclonal antibodies, some samples from patients who have been treated with monoclonal antibodies or have received them for diagnostic purposes can give erroneous findings. Human anti-mouse antibodies (HAMA) or heterophilic antibodies in human serum can react with the immunoglobulins included in the assay components causing interference and an anomalous result. Samples from patients routinely exposed to animals or animal serum products can demonstrate this type of interference. These reagents have been formulated to minimize the risk of interference; however, potential interactions between rare sera and test components can occur.”

### Interferences from Drugs and other Substances

The following studies were conducted based on CLSI EP7-A2 to test for various interfering drugs and other substances spiked individually into a human serum sample containing a single CEA concentration of 5 µg/L (5 ng/mL). Potential interference in the CEA assay is designed to be ≤ 10%. The average recovery was determined. It was



observed during the study to range between 91.6 and 108.4%.

Drug	Drug Concentration	Drug	Drug Concentration
Acetaminophen	300 µg/mL	Levodopa	20 µg/mL
Acetyl Cysteine	150 µg/mL	Lovastatin	2.5 µg/mL
Acetylsalicylic acid	500 µg/mL	Methyldopa	20 µg/mL
Ampicillin-sodium	1000 µg/mL	Metronidazole	200 µg/mL
Ascorbic Acid	300 µg/mL	Naprosyn sodium	500 µg/mL
Atrovastatin	3 µg/mL	Oxaliplatin	100 µg/mL
Cefoxitin	2500 µg/mL	Phenylbutazone	400 µg/mL
Cyclosporine A	10 µg/mL	Prednisone	5 µg/mL
Doxycycline hyclate	50 µg/mL	Rifampicin	60 µg/mL
Furosemide	4000 µg/mL	Tegafur with Uracil	50 µg/mL
Ibuprofen	1000 µg/mL	Theophylline	50 µg/mL
Irinotecan	100 µg/mL	Warfarin	50 µg/mL

*f. Assay cut-off:*

This is a test for serial monitoring. No cutoff for CEA monitoring has been recommended or defined even in the scientific literature. The user must choose their own percent change between two consecutive visits as their cutoff. The sponsor has provided in the package insert several example cutoffs with their attendant percent positive and negative agreement values as seen in the table below.

Percent Change Between Two Consecutive Visits	Percent Positive Agreement	Percent Negative Agreement	Sum of the Lower 95% Confidence Interval of Percent Positive and Negative Agreements
6.25%	57 %	57 %	92.9 %
15%	54 %	71 %	104.1 %
30%	47 %	89 %	117.5 %
44%	40 %	90 %	111.3 %

2. Comparison studies:

*a. Method comparison with predicate device:*

Study Design. Patient serum samples were used to compare the Olympus CEA assay on the Olympus AU3000i system against another commercially available CEA assay using CLSI protocol EP9-A2.<sup>15</sup> The patient serum samples were derived from apparently healthy individuals, various benign and malignant conditions along with colorectal serial sets. See expected values and reference ranges below for more details about the samples. Results using Deming regression analysis were as follows in micrograms per mL and ng/mL.

N	Range of concentrations	Intercept (95% CI)	Slope (95% CI)	Correlation Coefficient
1671	0.21 – 456.80	1.24 (0.98 – 1.51)	0.92 (0.87 – 0.97)	0.989

Three hundred (300) retrospective samples were obtained from sample banks obtained from 81 male and female colorectal cancer patients ranging in age from 8 to 98 years of age. Analysis using Deming Regression yielded the following in µg/L and ng/mL:

N	Range of concentrations	Intercept (95% Confidence Interval)	Slope (95% Confidence Interval)
300	0.99 – 3441	-3.28 (-5.54 - -1.09)	1.08 (1.02 – 1.14)

Note: This regression study included 5 samples above the highest calibrator that were diluted for assay.

*b. Matrix comparison:*

Study Design. Seventy-five (75) matched patient serum (Y) and Lithium heparin plasma samples (x) across the measurable range of the assay were used to compare serum vs. lithium heparin plasma using CLSI protocol EP9-A2.

Results:

N	Range of concentrations	Intercept	Slope	Correlation Coefficient
75	1.97 – 495.56	-0.12	0.99	0.99

Conclusion: Serum and Lithium heparin plasma samples are interchangeable across the assay range.

3. Clinical studies:

*a. Clinical Sensitivity:*

Three hundred (300) retrospective samples were obtained from sample banks obtained from 81 male and female colorectal cancer patients ranging in age from 8 to 98 years of age. Disease progression (or lack of progression) was determined by the subject's physician based on any or a composite of all of the following:

1. Examination of the patient for clinical signs and symptoms, including the results of laboratory tests that are current standard of care for the assessment of colorectal cancer disease status.
2. Examination of radiographic findings (imaging) ordered as standard of care that can be used for the assessment of colorectal cancer disease status. Radiographic findings include results from various imaging techniques such as Magnetic Resonance Imaging, Ultrasound, etc.
3. Interviews with the subject as to how the subject felt, any symptoms the subject experienced, and how the subject felt compared to previous time intervals.

An analysis of the percent change in Olympus CEA results between each of the 219 evaluable visit pairs was performed. The percent change is informative when the lower bound of the 95 % confidence interval of the sum of the percent positive agreement and percent negative agreement adds to > 100%. Presented below is a table of several percent changes and their corresponding percent positive and negative agreements as

examples of what might be expected with different percent changes seen with the test.

Percent Change Between Two Consecutive Visits	Percent Positive Agreement	Percent Negative Agreement	Sum of Percent Positive and Negative Agreements	Sum of the Lower 95% Confidence Interval of Percent Positive and Negative Agreements
6.25%	57 %	57 %	115%	92.9 %
15%	54 %	71 %	125%	104.1 %
30%	47 %	89 %	136%	117.5 %
44%	40 %	90 %	130%	111.3 %

For the Olympus CEA change of 30% between 2 consecutive visits, the performance of Olympus CEA assay for the 219 evaluable observation pairs is presented in the table below as an example.

	Progression	No progression	Total
<b>% Change &gt; 30 %</b>	27	26	53
<b>% Change ≤ 30 %</b>	30	136	166
<b>Total</b>	57	152	219

Percent positive agreement (equivalent to clinical sensitivity) measures the percentage of visits when the change in Olympus CEA value exceeds 30% compared to the CEA value at the previous visit and there is a corresponding disease progression at this visit. **The percent positive agreement is 47.4 % (27/57)** with a 95 % CI: 34.0 % to 61.0 %.

Percent negative agreement measures the percent of visits when a percent change in Olympus CEA value is less than 30% relative to the same measurement at the previous visit and there is a corresponding no disease progression (Responding, Stable, or No Evidence of Disease) at this visit. **The percent negative agreement is 89.5% (136/152)** with a 95 % CI: 83.5 % - 93.9 %.

*b. Clinical specificity:*

Three hundred (300) retrospective samples were obtained from sample banks obtained from 81 male and female colorectal cancer patients ranging in age from 8 to 98 years of age. Disease progression (or lack of progression) was determined by the subject's physician based on any or a composite of all of the following:

1. Examination of the patient for clinical signs and symptoms, including the results of laboratory tests that are current standard of care for the assessment of colorectal cancer disease status.
2. Examination of radiographic findings (imaging) ordered as standard of care that can be used for the assessment of colorectal cancer disease status. Radiographic findings include results from various imaging techniques such as Magnetic Resonance Imaging, Ultrasound, etc.
3. Interviews with the subject as to how the subject felt, any symptoms the subject experienced, and how the subject felt compared to previous time

intervals.

An analysis of the percent change in Olympus CEA results between each of the 219 evaluable visit pairs was performed. The percent change is informative when the lower bound of the 95 % confidence interval of the sum of the percent positive agreement and percent negative agreement add to > 100%.

Percent Change Between Two Consecutive Visits	Percent Positive Agreement	Percent Negative Agreement	Sum of Percent Positive and Negative Agreements	Sum of the Lower 95% Confidence Interval of Percent Positive and Negative Agreements
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% Change > 30 %	27	26	53
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Percent negative agreement (equivalent to specificity) measures the percent of visits when a percent change in Olympus CEA value is less than 30% relative to the same measurement at the previous visit and there is a corresponding no disease progression (Responding, Stable, or No Evidence of Disease) at this visit. **The percent negative agreement Presented below is a table of several percent changes and their corresponding percent positive and negative agreements as examples of what might be expected with different percent changes with the test is 89.5% (136/152) with a 95 % CI: 83.5 % - 93.9 %.**

*c. Other clinical supportive data (when a. and b. are not applicable):*

a. and b. are applicable.

4. Clinical cut-off:

This is a test for serial monitoring. No cutoff for CEA monitoring has been recommended or defined even in the scientific literature. The user must choose their own percent change between two consecutive visits as their cutoff. The sponsor has provided in the package insert several example cutoffs with their attendant percent positive and

negative agreement values as seen in the table below. The percent change is informative when the lower bound of the 95% confidence interval of the sum of the percent positive agreement plus the percent negative agreement adds to > 100%.

Percent Change Between Two Consecutive Visits	Percent Positive Agreement	Percent Negative Agreement	Sum of the Lower 95% Confidence Interval of Percent Positive and Negative Agreements
6.25%	57 %	57 %	92.9 %
15%	54 %	71 %	104.1 %
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44%	40 %	90 %	111.3 %

5. Expected values/Reference ranges:

A study using the Olympus CEA assay on 279 samples from self-reported apparently healthy non-smokers and 148 smokers (age18-60 years) with no history of malignancy other than non-invasive skin cancer gave the following results according to CLSI protocol C28-A2. Percentiles were determined non-parametrically.

	<b>Median</b> <b>[ng/mL],[μg/L]</b>	<b>97.5<sup>th</sup> Percentile</b> <b>[ng/mL],[μg/L]</b>
Non-smokers	2.39	5.64
Smokers	2.77	8.87

Expected values can vary with age, sex, sample type, diet and geographical location. Each laboratory should verify the transferability of the expected values to its own population and if necessary determine its own reference range following established procedures such as CLSI procedure C28-A2.<sup>10</sup>

In the table below a breakdown of CEA results from apparently healthy individuals, along with patients with a variety of both benign and malignant conditions are presented.

	Number of Subjects (n)	n (%)				
		0.0 – 5.0 [μg/L],[ng/mL]	5.1 – 10.0 [μg/L],[ng/mL]	10.1 – 50 [μg/L],[ng/mL]	51 – 500 [μg/L],[ng/mL]	>500 [μg/L],[ng/mL]
<b>Apparently Healthy</b>						
Non-smokers	279	270 (96.8%)	8 (2.9%)	1 (0.4%)	-	-
Smokers	148	132 (89.2%)	15 (10.1%)	1 (0.7%)	-	-
<b>Benign Conditions</b>						
Prostate/Testicular	85	76 (89.4%)	8 (9.4%)	1 (1.2%)	-	-
GI tract/Lung	109	103	5 (4.6%)	1 (0.9%)	-	-

		(94.5%)				
Diabetes	104	89 (85.6%)	13 (12.5%)	2 (1.9%)	-	-
Heart/Liver	109	98 (89.9%)	10 (9.2%)	1 (0.9%)	-	-
Breast	46	41 (89.1%)	5 (10.9%)	-	-	-
<b>Malignant Conditions</b>						
Lung (treated)	85	48 (56.5%)	22 (25.9%)	7 (8.2%)	6 (7.0%)	2 (2.4%)
Liver (treated)	25	17 (68.0%)	5 (20.0%)	3 (12.0%)	-	-
GI tract (treated)	57	38 (66.7%)	9 (15.8%)	9 (15.8%)	1 (1.8%)	-
Prostate/Testicular/ Bladder (treated)	131	117 (89.3%)	13 (9.9%)	1 (0.8%)	-	-
Colorectal*	146	77 (52.7%)	18 (12.3%)	27 (18.5%)	19(13.0%)	5 (3.4%)
Breast (treated)	55	33 (60.0%)	11 (20.0%)	10 (18.2%)	1 (1.8%)	-

\*Mixed cohort of treated and untreated patients.

**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.