

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

A. 510(k) Number:

K083464

B. Purpose for Submission:

To determine substantial equivalence for the Premier™ CAMPY Enzyme Immunoassay (EIA) used to detect *Campylobacter* (*C. jejuni* and *C. coli*) antigens

C. Measurand:

Campylobacter jejuni and *Campylobacter coli* antigens

D. Type of Test:

Enzyme Immunoassay

E. Applicant:

Meridian Bioscience, Inc.

F. Proprietary and Established Names:

Premier™ CAMPY Enzyme Immunoassay (EIA)

G. Regulatory Information:

1. Regulation section:

21 CFR § 866.3110, *Campylobacter* spp

2. Classification:

Class I

3. Product code:

LQP - *Campylobacter* spp.

4. Panel:

Microbiology (83)

H. Intended Use:

1. Intended use:

Premier™ CAMPY enzyme immunoassay (EIA) is an in vitro qualitative procedure for the detection of specific *Campylobacter* antigens in stool samples from patients with signs and symptoms of gastroenteritis. Premier CAMPY detects *C. jejuni* and *C. coli* in human stool that may be either unpreserved or preserved in Cary Blair-based transport media. Test results are to be used in conjunction with information obtained from the patient's clinical evaluation and other diagnostic procedures.

Premier CAMPY is intended for use in hospital, reference or state laboratory settings. The device is not intended for point-of-care use.

2. Indications for use:

Premier™ CAMPY enzyme immunoassay (EIA) is an in vitro qualitative procedure for the detection of specific *Campylobacter* antigens in stool samples from patients with signs and symptoms of gastroenteritis. Premier CAMPY detects *C. jejuni* and *C. coli* in human stool that may be either unpreserved or preserved in Cary Blair-based transport media. Test results are to be used in conjunction with information obtained from the patient's clinical evaluation and other diagnostic procedures.

Premier CAMPY is intended for use in hospital, reference or state laboratory settings. The device is not intended for point-of-care use.

3. Special conditions for use statement(s):

For Prescription Use Only

4. Special instrument requirements:

None

I. Device Description:

Premier CAMPY is an in vitro diagnostic, microwell-based, enzyme-linked immunoassay for the detection of common antigens found on *Campylobacter jejuni* and *C. coli* in stool samples from patients with signs and symptoms of Campylobacteriosis.

The assay kit contains the following components:

1. Premier CAMPY Microwells: Monoclonal antibody–coated microwells. The antibodies are specific to *C. jejuni* and *C. coli*
2. Premier CAMPY Enzyme Conjugate: HRP-conjugated monoclonal antibodies specific to *C. jejuni* and *C. coli* in a buffered protein solution containing 0.1% ProClin® and 0.03% gentamicin as preservatives
3. Premier 20X Wash Buffer III: Concentrated wash buffer containing 0.1% ProClin® and 0.3% gentamicin as preservatives
4. Premier Substrate I: Buffered solution containing urea peroxide and tetramethylbenzidine at pH 5.0
5. Premier Stop Solution I: 1M Phosphoric acid
6. Premier CAMPY Sample Diluent/Negative Control: Buffered protein solution containing 0.1% ProClin® and 0.03% gentamicin as preservatives.
7. Premier CAMPY Positive Control: Inactivated *C. jejuni* in a buffered protein solution containing 0.094% sodium azide and 0.03% gentamicin as preservatives
8. Transfer pipettes
9. Microwell strip holder
10. Microwell plate sealers

No calibrators are used with this device.

J. Substantial Equivalence Information:

1. Predicate device name(s):
ProSpecT Campylobacter EIA
2. Predicate 510(k) number(s):
K982315

3. Comparison with predicate:

Similarities and Differences

<i>Item</i>	<i>Premier CAMPY</i>	<i>Predicate Device ProSpecT Campylobacter</i>
Assay type	EIA	EIA
Intended use		
Qualitative/Quantitative	Qualitative	Qualitative
Screening, diagnostic or identification test	Diagnostic	Diagnostic
Calibrator	No	No
Monitoring therapy	No	No
Reagents/components		
Microwells	Yes	Yes
Sample Diluent	Yes	Yes
Enzyme Conjugate	Yes	Yes
Wash Buffer	Yes	Yes
Substrate	Yes	Yes
Stop Solution	Yes	Yes
Positive Control	Yes	Yes
Negative Control	Yes	Yes
Species detected		
<i>C. jejuni</i>	Yes	Yes
<i>C. coli</i>	Yes	Unk
<i>C. lari</i>	No	No
<i>C. fetus</i>	No	No
Reading method		
Visual	Yes	Yes
Spectrophotometric	Yes	Yes
End point	Pos = definite yellow color Neg = Colorless to very faint yellow	Pos = yellow color Negative = Colorless
Calibrator	No	No
Equipment	General laboratory semiautomated washer (optional) General laboratory spectrophotometer (optional)	General laboratory semiautomated washer (optional) General laboratory spectrophotometer (optional) StatFax microplate incubator/shaker (optional)

<i>Item</i>	<i>Premier CAMPY</i>	<i>Predicate Device ProSpecT Campylobacter</i>
<i>Antibody sources</i>		
Solid phase (microplate)	Mouse monoclonal	Rabbit polyclonal
Enzyme conjugate	Mouse monoclonal	Rabbit polyclonal
<i>Sample Types</i>		
Human stool (direct)	Yes	Yes
Broth culture	No	Yes
<i>Endpoint determinations</i>		
Positive (dual wavelength)	Yes ≥ 0.100	Yes ≥ 0.140
Negative (dual wavelength)	Yes < 0.100	Yes < 0.100
Indeterminant (dual wavelength)	None	Yes 0.100 to 0.139

Differences include Specific Detection of *C. coli* and Antibody Source.

The Premier™ CAMPY EIA kit can specifically detect *C. coli* and includes anti-mouse monoclonal antibodies, while the predicate (K982315- ProSpecT Campylobacter EIA) cannot distinguish between *C. coli* and *C. jejuni* and contains anti-rabbit polyclonal antibodies.

K. Standard/Guidance Document Referenced:

EPI2-A2, Vol. 28, No. 3. User protocol for evaluation of qualitative test performance, approved guideline. 2nd ed., CLSI (Section 8.3.1)

L. Test Principle:

Premier CAMPY is an enzyme immunoassay for the direct detection of Campylobacter antigens in human stool samples. Breakaway microwells are coated with Campylobacter-specific monoclonal antibodies. Diluted patient specimen is added to the microwells and incubated. Upon completion of the incubation, a wash step is performed to remove unbound material and a Horseradish Peroxidase (HRP)–Anti–Campylobacter conjugate is added to the washed microwells. If Campylobacter antigens are present, an antibody-enzyme complex is formed. A second wash step is performed to remove unbound materials and a chromogen substrate is added to the microwells. A blue color develops in the presence of bound enzyme. Premier Stop

Solution I is added, changing the initial blue reaction to yellow. Test results are interpreted visually or spectrophotometrically.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Assay precision, intra-assay variability and inter-assay variability were assessed with a reference panel prepared from moderate positive (n =2), negative (n =2), high negative (n =3) and low positive (n = 3) samples. High negative, low positive and moderate positive samples were prepared by inoculating negative stool matrix with known quantities of *C. jejuni*. In the case of low positive and high negative samples, the inoculum was added at concentrations that were at, or just below, the assay LoD. Aliquots of each panel were tested for five days, twice each day at three different test sites (Sites A, B and C). At least two technologists performed testing at each site. The expected results were obtained 100% of the time.

The sponsor's pre-determined acceptance criteria were as follows:

- Premier™ Campylobacter and culture will result in performance values of 90% sensitive and 90% specific \pm 10%, at 90% confidence
- Fewer than 10% false positive results
- Fewer than 10% false negative

b. Linearity/Assay Reportable Range

N/A

c. Traceability, Stability, Expected Values (controls, calibrators or methods)

Stability

To validate labeling claims regarding the use of preserved and/or frozen samples, the investigator subjected a panel of specimens, preserved in Cary Blair-based transport media, to two freeze-thaw cycles (at -20°C and 2-8°C). The sample panel included low positive and high negative samples prepared by spiking negative matrices with *C. jejuni*.

The sponsor's interpretation of results for Premier CAMPY at OD_{450/630} was as follows:

Negative < 0.100

Positive \geq 0.100

These results suggest that stool samples are stable whether preserved or frozen; thus, effectiveness of *C. jejuni* and *C. coli* detection in such samples is not compromised.

Human stool samples, unpreserved: Samples should be received in an airtight transport container and stored at 2–8⁰ C prior to testing. Samples should be tested as soon as possible, but may be held up to 96 hours at 2–8⁰ C. Samples that will not be tested within 96 hours should be frozen immediately upon receipt and stored at ≤ –20 C until tested. Specimens may be frozen and thawed twice.

Human stool samples, preserved in Cary Blair-based media: Samples should be stored at 2–8⁰ C prior to testing. Samples should be tested as soon as possible, but may be held up to 96 hours at 2–8 C⁰. Samples that will not be tested within 96 hours should be frozen immediately upon receipt and stored at ≤ –20 C until tested. Specimens may be frozen and thawed twice.

Controls: The controls are used to monitor reagent reactivity. Failure of the controls to produce the expected results can mean that one or more of the reagents are no longer reactive at the time of use that the test was not performed correctly, or that reagents or samples were not added. Repeat the control tests as the first step in determining the root cause of the failure.

d. *Detection limit:*

The following tables summarize the study design used to determine the Limit of Detection (LoD), for the Premier™ CAMPY kit, of *C. jejuni* and *C. coli*, respectively. The analytical sensitivity of these assays for *C. jejuni* and *C. coli* were based on 45 tests for each measurand and with a stated 95 % probability of obtaining positive responses (i.e., LoD) at the following levels of the measurands:

C. jejuni - 1.2 x 10⁶ cells/mL
C. coli - 8.0 x 10⁶ cells/ml

Measurand: *Campylobacter coli*

	Sample #1 (2 dil. below LoB)		Sample #2 (1 dil. below LoB)		Sample #3 (LoB)		Sample #4 (1 dil. above LoB)		Sample #5 (2 dil. above LoB)	
Organism concentration	1.0×10^6		2.0×10^6		4.0×10^6		8.0×10^6		1.6×10^7	
Kit Lot Number	# Pos	# Neg	# Pos	# Neg	# Pos	# Neg	# Pos	# Neg	# Pos	# Neg
Lot #1	0	15	0	15	0	15	15	0	15	0
Lot #2	0	15	0	15	12	3	15	0	15	0
Lot #3	0	15	0	15	8	7	15	0	15	0
Total (for 3 lots)	0	45	0	45	20	25	45	0	45	0
Meets requirement for final LoB? (yes/no)	No		No		Yes		No		No	
Meets requirement for LoD? (yes/no)	No		No		No		Yes		No	

Measurand: *Campylobacter jejuni*

	Sample #1 (2 dil. below LoB)		Sample #2 (1 dil. below LoB)		Sample #3 (LoB)		Sample #4 (1 dil. above LoB)		Sample #5 (2 dil. above LoB)	
Organism concentration	1.5×10^5		3.0×10^5		6.5×10^5		1.2×10^6		2.4×10^6	
Kit Lot Number	# Pos	# Neg	# Pos	# Neg	# Pos	# Neg	# Pos	# Neg	# Pos	# Neg
Lot #1	0	15	0	15	0	15	15	0	15	0
Lot #2	0	15	0	15	15	0	15	0	15	0
Lot #3	0	15	0	15	12	3	15	0	15	0
Total (for 3 lots)	0	45	0	45	27	18	45	0	45	0
Meets requirement for final LoB? (yes/no)	No		No		Yes		No		No	
Meets requirement for LoD? (yes/no)	No		No		No		Yes		No	

e. Analytical specificity:

Interference Testing

In this study, the sponsor added selected drugs, and other non-microbial substances that might be present in stool samples from healthy persons or patients with signs and symptoms of gastroenteritis, to three positive and three negative samples. They then inoculated the samples with *C. jejuni* near the limit of detection (LoD) of the assay. The final concentrations of the substances in the samples were as follows:

- Barium Sulfate (5 mg/mL)
- Fecal fat (equivalent to 2.65 mg stearic plus 1.3 mg palmitic acids per mL)
- Hemoglobin (as methhemoglobin) (3.2 mg/mL)
- Imodium AD® (0.00667 mg/mL)
- Kaopectate® (0.87 mg/mL)
- Mucin (3.33 mg/mL)
- Mylanta® (4.2 mg/mL), Pepto-Bismol® (0.87 mg/mL)
- Prilosec® (0.5 mg/mL)
- Tagamet® (0.5 mg/mL)
- TUMS® (0.5 mg/mL), whole blood (5% v/v)

Meridian Biosciences tested the spiked samples in parallel with an unspiked dilution control for reference. All samples were tested in triplicate. The sponsor provides the following interpretation of results for Premier CAMPY at OD_{450/630}:

Negative test: <0.100

Positive Test: ≥ 0.100

The negative results observed suggest that none of the potentially interfering substances

meets the criteria for an interferent.

Cross-reactivity Study

Microorganisms that were present as normal intestinal flora or associated with gastroenteritis were evaluated as to their effects on assay performance. Fungi and bacteria were tested at final concentrations in human stool of 1.1×10^8 CFU/mL. Viruses were tested at final concentrations of 1.3×10^4 to 3.1×10^6 TCID₅₀/mL. The sponsor provides the following interpretation of results for Premier CAMPY at OD_{450/630}:

Negative test: <0.100

Positive Test: ≥ 0.100

No cross-reactivity was observed.

These results suggest that none of the following organisms in stool reacts with Premier CAMPY.

Normal/Gastritis Associated Intestinal Flora		
Bacteria	Fungi	Viruses
<i>Aeromonas hydrophila</i> <i>Bacteroides fragilis</i> <i>Campylobacter fetus</i> <i>C. lari</i> <i>Clostridium difficile</i> <i>C. perfringens</i> <i>Enterobacter cloacae</i> <i>Enterococcus faecalis</i> <i>Escherichia coli</i> <i>E. coli</i> 0157:H7 <i>E. fergusonii</i> <i>E. hermannii</i> <i>Helicobacter pylori</i> <i>Klebsiella pneumoniae</i> <i>Lactococcus lactis</i> <i>Listeria monocytogenes</i> <i>Peptostreptococcus anaerobius</i> <i>Plesiomonas shigelloides</i> <i>Proteus vulgaris</i> <i>Pseudomonas aeruginosa</i> <i>P. fluorescens</i> <i>Salmonella</i> Groups BE <i>Serratia liquefacie</i> <i>S. marcescens</i> <i>Shigella boydii</i> <i>S. dysenteriae</i> <i>S. Flexneri</i> <i>S. sonnei</i> , <i>Staphylococcus aureus</i> <i>S. epidermidis</i> <i>Vibrio parahaemolyticus</i> <i>Yersinia enterocolitica</i>	<i>Candida albicans</i>	Adenovirus Types 40 and 41 Coxsackievirus Echovirus Rotavirus

Strain Reactivity Study

The sponsor tested the reactivity of the following *Campylobacter* stock cultures from different sources, at 1.1×10^8 CFU/mL with Premier CAMPY: *C. coli* strains 10956, 17755, 36994 and 53138 and *C. jejuni* strains 6951, 10940, 12081, 29411 and 38106. In this study, triplicates of each strain were spiked into a negative stool matrix. The negative sample matrix was confirmed nonreactive with the predicate device. The control was prepared by spiking the matrix with buffer instead of organism. Results

showed that the negative control was nonreactive when tested, while each of the stool specimens spiked with the above listed *Campylobacter* stock cultures was reactive.

e. Assay cut-off:

Interpretation of Results

Visual Reading

Negative = Colorless to very faint yellow

Positive = Definite yellow color

A very faint yellow color must be evaluated by a spectrophotometric reading.

Spectrophotometric Single Wavelength (450 nm)

Negative: < 0.150

Positive: ≥ 0.150

Negative Control: < 0.150

Positive Control: ≥ 0.600

Spectrophotometric Dual Wavelength (450/630 nm)

Negative: < 0.100

Positive: ≥ 0.100

Negative Control: < 0.100

Positive Control: ≥ 0.600

If a Negative Control is < 0.000 , blank the plate reader again to air and reread the plate.

A positive result indicates the presence of *Campylobacter* antigens. A negative result indicates the absence of *Campylobacter* antigens, or that the level of antigens is below what can be detected by the assay. The magnitude of the OD above the cutoff is neither indicative of the severity or extent of *Campylobacter* infection, nor can it be correlated to an endpoint titer. Extremely strong positive samples may yield either an intense yellow color or a purple precipitate within a few minutes of stopping the reaction. In this case, the spectrophotometer may yield an “out” reading. This reading is considered a positive.

If the frequency of low positive results (OD between 0.150 and 0.200 for single wavelength and between 0.100 and 0.150 with dual wavelength) is greater than 5% of specimens tested, this may indicate insufficient washing, more, vigorous washing or increasing the washes to seven washes in step 5 of the procedure is recommended.

1. Comparison studies:

a. Method Comparison with Predicate:

N/A

b. Matrix comparison:

Specimen matrix interference has not been observed in this assay as samples are significantly diluted before testing in Sample Diluent. For this reason, the Positive and Negative Controls supplied as part of this assay are prepared in matrices similar to the Sample Diluent. If control materials that are identical in composition to test samples are preferred, the user can prepare these by diluting known positive and negative specimens in Sample Diluent according to the SPECIMEN PREPARATION section of the package insert.

3. Clinical studies:

a. Clinical Sensitivity

Meridian Bioscience established the performance of Premier CAMPY in clinical trials using bacterial culture as the reference comparator method. They conducted their clinical studies at four independent test sites located in the Western, Midwestern and Southeastern regions of the United States. In total, they tested 2073 qualified patient samples. Of these, 166 were retrospective frozen samples and the remaining 1907 prospective. The sponsor collected the majority (1862/2073) of samples in a Cary Blair-based transport and preservative medium. They tested the remaining 211 samples in the unpreserved state. They collected stool from both males (41 %) and females (57%); however, they were unaware of the sex of 2% of their patients. The age groups of the patients ranged from less than one month of age to 97 years. No differences in test performance were observed based on patient age or sex. The following tables illustrate the assay performance by clinical site.

Table 3 (Suppl) Combined data – stratified by test site

	Premier CAMPY			ProSpecT Campylobacter			
Site #1							
Bacterial Culture	Positive	Negative	Total	Positive	Negative	Indeterminate	Total
Positive	22	1	23	22	1	0	23
Negative	4	189	193	0	193	0	193
Total	26	190	216	22	194	0	216
			95% CI			95% CI	
Sensitivity	22/23	95.7	79.0 – 99.2	22/23	95.7	79.0 – 99.2	
Specificity	189/193	97.9	94.8 – 99.2	193/193	100.0	98.0 – 100.0	
Correlation	211/216	97.7	94.7 – 99.0	215/216	99.5	97.4 – 99.9	
Site #2							
Bacterial Culture	Positive	Negative	Total	Positive	Negative	Indeterminate	Total
Positive	0	0	0	0	0	0	0
Negative	0	51	51	0	51	0	51
Total	0	51	51	0	51	0	51
			95% CI			95% CI	
Sensitivity	0/0	N/A	N/A	0/0	N/A	N/A	
Specificity	51/51	100	93.0 – 100.0	51/51	100	93.0 – 100.0	
Correlation	51/51	100	93.0 – 100.0	51/51	100	93.0 – 100.0	
Site 3							
Bacterial Culture	Positive	Negative	Total	Positive	Negative	Indeterminate	Total
Positive	26	1	27	24	3	0	27
Negative	82	1429	1511	36	1462	13	1511
Total	108	1430	1538	60	1465	13	1538
			95% CI			95% CI	
Sensitivity	26/27	96.3	81.7 – 99.3	24/27	88.9	71.9 – 96.1	
Specificity	1429/1511	94.6	93.3 – 95.6	1475/1511	96.8	95.7 – 97.5	
Correlation	1455/1538	94.6	93.4 – 95.6	1486/1538	96.6	95.6 – 97.4	
Site 4							
Bacterial Culture	Positive	Negative	Total	Positive	Negative	Indeterminate	Total
Positive	11	0	11	11	0	0	11
Negative	2	255	257	2	255	0	257
Total	13	255	268	13	255	0	268
			95% CI			95% CI	
Sensitivity	11/11	100.0	74.1 – 100.0	11/11	100.0	74.1 – 100.0	
Specificity	255/257	99.2	97.2 – 99.8	255/257	99.2	97.2 – 99.8	
Correlation	266/268	99.3	97.3 – 99.8	266/268	99.3	97.3 – 99.8	

b. Clinical Specificity

See 3a above

c. Other Clinical Supportive data

N/A

4. Clinical cut-off:

N/A

5. Expected Values/Reference Range:

The performance of Premier CAMPY was evaluated during 2008 in several

geographic regions of the United States. The incidence of positive samples (*C. jejuni* or *C. coli*) during this period was approximately 1.5%. The expected frequency for an individual laboratory may differ from this number since it is dependent on factors such as locale, time of the year, and whether an outbreak has occurred.

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.