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

A. 510(k) Number:

K162620

B. Purpose for Submission:

To obtain a substantial equivalence determination for Remel Spectra ESBL

C. Measurand:

Extended Spectrum β-Lactamase (ESBL) producing *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella oxytoca* and *Proteus mirabilis*

D. Type of Test:

Selective and differential culture medium

E. Applicant:

Remel, Inc.

F. Proprietary and Established Names:

Remel Spectra ESBL

G. Regulatory Information:

1. Regulation section:

   21 CFR 866.1700

2. Classification:

   II

3. Product code:

   JSO: Culture Media, Antimicrobial Susceptibility Test, Excluding Mueller Hinton Agar

4. Panel:

   83 Microbiology

H. Intended Use:

1. Intended use(s):

   Remel Spectra ESBL is a selective and differential growth medium for use in primary isolation and presumptive identification of Extended Spectrum β Lactamase (ESBL)-producing *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella oxytoca* and *Proteus mirabilis* to aid in prevention and control of these bacteria in a healthcare setting. Testing may be performed from either perirectal swabs or fresh stool specimens. Remel Spectra ESBL is not intended to diagnose ESBL infection, or to guide or monitor treatment.

   Do not report Spectra ESBL positive screening results. Subculture of presumptive positive colonies to non-selective medium (e.g., Tryptic Soy Agar with 5% sheep blood) is required for organism identification, confirmatory testing for ESBL, susceptibility testing and epidemiological typing.

   A lack of growth or the absence of pink, blue-turquoise-green or tan colonies on Spectra ESBL does not preclude the presence of ESBL producing organisms.

2. Indication(s) for use:

   Same as Intended Use

3. Special conditions for use statement(s):

   For prescription use only

   Do not report positive screening results obtained on Spectra ESBL. Subculture is required for organism identification and ESBL confirmatory testing.

   Colony color on Remel Spectra ESBL should not be used as a basis for organism identification. Colony color may vary based on the species, strain of organism, specimen, matrix and density of inoculum. While Spectra ESBL is a screening medium for ESBL-producing *E. coli*, *K. pneumoniae*, *K. oxytoca* and *P. mirabilis*, other *Enterobacteriaceae* that do not have CLSI interpretive screening guidelines for ESBL production may also grow on Spectra ESBL. It is important to subculture isolated pink, blue-turquoise-green or tan colonies from Spectra ESBL to non-selective medium to confirm identity, susceptibility and ESBL phenotype when screening for ESBL-producing microorganisms.

   Areas of heavy stool inoculum in the 1st or 2nd quadrants may cause ESBL producing strains of the targeted species to produce colony colors other than those expected. Only sample well isolated pink, blue-turquoise-green or tan colonies from the 3rd or 4th quadrants.

   A lack of growth or the absence of pink, blue-turquoise-green or tan colonies on Spectra

ESBL does not preclude the presence of ESBL-producing organisms. False negative results may occur due to variations in sampling, slow development or failure to develop the expected colony color, or the presence of organisms that are susceptible to the antimicrobial agents included in the Spectra ESBL medium.

When present at high concentration, some non-ESBL producing species may produce colonies that are consistent in appearance with those of ESBL-producing strains and suppress growth of target organisms.

The performance of Spectra ESBL has been established with fresh stool specimens and perirectal swabs only. Performance characteristics with rectal swabs or other specimens have not been established.

4. Special instrument requirements:

   None

I. Device Description:

Remel Spectra ESBL is a selective and differential culture medium that contains a combination of antibacterial agents that suppress the growth of non-Extended Spectrum β-Lactamase (non-ESBL) producing organisms and enable recovery of ESBL-producing strains of *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella oxytoca* and *Proteus mirabilis* from stool and perirectal swab specimens. The medium contains peptones that supply amino acids and essential nutrients to promote the growth of enteric Gram-negative organisms. Sodium chloride is present as a source of electrolytes and to maintain osmotic equilibrium. Metabolism of chromogenic substrates and other components of the medium enable visual differentiation of colonies of target and non-target organisms.

J. Substantial Equivalence Information:

1. Predicate device name(s):

   HardyCHROM ESBL

2. Predicate 510(k) number(s):

   K160512

3. Comparison with predicate:

Similarities		
Item	**Device**	**Predicate**
	Remel Spectra ESBL (K162620)	**HardyCHROM ESBL (K160512)**
Regulation	21 CFR 866.1700	Same
Product Code	JSO	Same
Device Class	II	Same
Intended Use	Remel Spectra ESBL is a selective and differential growth medium for use in primary isolation and presumptive identification of Extended Spectrum β Lactamase (ESBL)-producing *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella oxytoca* and *Proteus mirabilis* to aid in prevention and control of these bacteria in a healthcare setting. Testing may be performed from either perirectal swabs or fresh stool specimens. Remel Spectra ESBL is not intended to diagnose ESBL infection, or to guide or monitor treatment. Do not report Spectra ESBL positive screening results. Subculture of presumptive positive colonies to non-selective medium (e.g., Tryptic Soy Agar with 5% sheep blood) is required for organism identification, confirmatory testing for ESBL, susceptibility testing and epidemiological typing. A lack of growth or the absence of pink, blue-turquoise-green or tan	HardyCHROM ESBL is a selective and differential chromogenic medium which is intended for the qualitative and presumptive detection from stool specimens of: 1) *Enterobacteriaceae* that are potentially non-susceptible to ceftazidime and cefpodoxime; and 2) Extended-spectrum beta-lactamase (ESBL)-producing *Escherichia coli*, *Klebsiella pneumoniae* and *Klebsiella oxytoca*. The test is performed on stool specimens from patients at risk of harboring *Enterobacteriaceae* that are non-susceptible to 3rd generation cephalosporins or ESBL-producing *E. coli*, *K. pneumoniae* and *K. oxytoca*, and is intended as an aid in the detection, identification of colonization and control of these bacteria in a healthcare setting. HardyCHROM ESBL is not intended to diagnose infection or to guide or monitor treatment for infections. Results can be interpreted after incubation

Similarities		
Item	**Device**	**Predicate**
	Remel Spectra ESBL (K162620)	**HardyCHROM ESBL (K160512)**
	colonies on Spectra ESBL does not preclude the presence of ESBL producing organisms.	for 18-24 hours. Subculture to non-selective medium is required for confirming identification, antimicrobial susceptibility testing and epidemiological typing. A lack of growth or the absence of pink, blue or yellow/gold colonies on HardyCHROM ESBL does not preclude the presence of *Enterobacteriaceae* that are non-susceptible to 3rd generation cephalosporins or ESBL producing organisms.
Specimen Type	Stool or perirectal swabs	Stool
Method of Inoculation	Direct specimen	Same
Method of Interpretation	Manual, visual	Same
Test Methodology	Growth of target organisms in the presence of selective agents	Same
Organism Identification and Antimicrobial Susceptibility	Presumptive – confirmation of organism identity and ESBL phenotype required	Same
Method of Colony Differentiation	Metabolism of chromogenic and other substrates	Same
Incubation	Aerobic; 18-24 hours	Same
Subculture	Required for confirmation of identity and ESBL phenotype	Same

Differences		
Item	**Device**	**Predicate**
	Remel Spectra ESBL (K162620)	**HardyCHROM ESBL (K160512)**
Organisms Detected	ESBL producing *E. coli*, *K. pneumoniae*, *K. oxytoca* and *P. mirabilis*	*Enterobacteriaceae* that are potentially non-susceptible to ceftazidime and cefpodoxime

Differences		
Item	**Device**	**Predicate**
	Remel Spectra ESBL (K162620)	**HardyCHROM ESBL (K160512)**
		ESBL producing *E. coli*, *K. pneumoniae* and *K. oxytoca*
Reference Method	Inoculation of MacConkey agar with a 10µg cefpodoxime disc between the 1st and 2nd quadrants, followed by selection of isolated colonies from within the zone of inhibition, microbial identification and susceptibility testing for confirmation of ESBL phenotype.	Growth in Trypticase Soy enrichment broth with ceftazidime or cefotaxime, followed by subculture to MacConkey agar with identification and antimicrobial susceptibility testing of isolated colonies for confirmation of ESBL phenotype.

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

CLSI. Performance Standards for Antimicrobial Susceptibility Testing. 23rd ed. CLSI supplement M100S. Wayne, PA: Clinical and Laboratory Standards Institute; 2013.

CLSI. Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Approved Standard. 9th ed. CLSI M07-A9. Wayne, PA. Clinical and Laboratory Standards Institute; 2012.

L. Test Principle:

Remel Spectra ESBL contains a combination of antibacterial agents that inhibit non-ESBL producing *Enterbacteriaceae* and suppress the growth of most organisms that carry AmpC β-lactamases, as well as other non-ESBL producing flora. Peptones supply amino acids and essential nutrients which promote the growth of enteric gram-negative bacteria. Sodium chloride is a source of essential electrolytes and maintains osmotic equilibrium. A chromogenic mixture enables differentiation of isolates that express galactosidase (turquoise-green colonies) and/or glucuronidase (blue or pink colonies). Hydrolysis of tryptophan also produces tan colonies with a brown halo.

Pink, blue-turquoise-green or tan colonies are presumptively positive for ESBL-producing *E. coli*, *K. pneumoniae*, *K. oxytoca*, or *P. mirabilis.* Subculture is required for organism identification and ESBL confirmatory testing.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

The reproducibility of Spectra ESBL was evaluated using a blinded nine member panel of isolates comprised of seven ESBL producing strains of *E. coli* (3), *Klebsiella pneumoniae* (3), and *K. oxytoca* (1) and one strain each of non-ESBL producing *E. coli* and *K. pneumoniae*. Testing was performed by 2 operators at 3 sites on each of 5 days using two levels of each panel member (approximately 10^4 and 10^6 CFU/mL) (2 operators x 5 days x 3 sites x 9 panel members = 270 results per organism level). At both target levels, there was 100% agreement with expected colony color for all the ESBL-producing organisms (210/210) and there were no false positive results with either of the non-ESBL-producing strains (0/60). The results of the study were acceptable.

b. Linearity/assay reportable range:

Not Applicable

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

Weekly Quality Control testing was performed at each of the sites that participated in the Clinical Study described in **Section M(3)**. The Quality Control strains used in the study are shown in **Table 1**. During the course of the Clinical Study, each control strain was tested a total of 50 times and there were no unexpected results (100% agreement). These results are acceptable.

Table 1. Quality Control strains used in the Clinical Study

Species	Strain	β-Lactamase Genotype	Expected Colony Color or Result
E. coli	ATCC BAA-201	TEM-3	Blue-Turquoise-Green
K. pneumoniae	ATCC 700603	SHV-18	Blue-Turquoise-Green
P. mirabilis	511-116	Not known	Tan
E. coli	ATCC 25922	Not applicable	No Growth

ATCC: American Type Culture Collection

d. Detection limit:

Limit of Detection
The Limit of Detection (LOD) of Spectra ESBL was determined by plating serial dilutions of 11 confirmed ESBL-producing strains of bacteria that were prepared in saline (100µL per plate). The LOD was also determined for a subset of strains suspended in a liquefied stool matrix using an inoculum volume of 1mL per plate. The results of both these studies are summarized in **Table 2**. The LOD in saline

ranged from 1-1000 CFU/mL, depending on the strain. For each of the 4 strains tested in stool matrix, the LOD was 10 CFU/mL.

Table 2. Summary of the results of the LOD Study for Spectra ESBL

Species	Strain	β-Lactamase Genotype	LOD (CFU/mL)	
			Saline [1]	Stool [2]
E. coli	ATCC BAA-201	TEM-3	10	10
	ATCC BAA-196	TEM-10	1	Not tested
	ATCC BAA-198	TEM-26	10	Not tested
	ATCC BAA-200	SHV-4	1	Not tested
	4	Not known	10	Not tested
	32	Not known	1000	Not tested
	UK RDCC 0223	CTX-M14	Not tested	10
K. pneumoniae	ATCC 700603	SHV-18	100	10
	5	Not known	100	Not tested
	17	Not known	100	Not tested
K. oxytoca	ATCC 51983	SHV-5	1	10
P. mirabilis	511-116	Not known	10	Not tested

[1] Lowest concentration at which both replicates yielded colonies of the expected color
[2] Lowest concentration at which all 5 replicates yielded colonies of the expected color

Inclusivity
The ability of Spectra ESBL to support growth of ESBL producing organisms was evaluated by testing a total of 48 confirmed ESBL-producing strains: *E. coli* (27), *K. pneumoniae* (8), *K. oxytoca* (7) and *P. mirabilis* (6). All of the strains grew with the expected colony color when diluted in saline and inoculated at a concentration of 10^3 CFU/plate.

e. *Analytical specificity:*

The analytical specificity of Spectra ESBL was evaluated by testing a panel of 61 confirmed non-ESBL producing strains of *Enterobacteriaceae* as well as 49 strains of non-*Enterobacteriaceae* species at an inoculum concentration of 10^4 CFU/plate. The organisms tested are summarized in **Tables 3** and **4**.

Table 3. Non-ESBL producing *Enterobacteriaceae* tested in the Spectra ESBL Analytical Specificity Study

Non-ESBL Producing *Enterobacteriaceae*	Number of Strains Tested	Pink/Blue-Turquoise-Green/Tan Colonies on ESBL Spectra	
		Negative	Positive [1]
Citrobacter freundii	1	0	1
Enterobacter aerogenes	1	0	1
Enterobacter cloacae	8	1	7
Escherichia coli	23	23	0
Klebsiella oxytoca	1	0	1
Klebsiella pneumoniae	11	3	8 [2]
Providencia stuartii	1	1	0
Salmonella spp.	4	4	0
Serratia marcescens	2	2	0
Shigella boydii	1	1	0
Shigella dysenteriae	1	1	0
Shigella flexneri	1	1	0
Shigella sonnei	3	3	0
Yersinia enterocolitica	2	2	0
Yersinia kristensenii	1	1	0

[1] 17/18 strains that exhibited pink/blue-turquoise-green/tan colonies on Spectra ESBL were non-susceptible to ceftazidime and cefotaxime
[2] 1 strain of *K. pneumoniae* was susceptible to both ceftazidime and cefotaxime

Table 4. Non-*Enterobacteriaceae* tested in the Spectra ESBL Analytical Specificity Study

Species Name	Number of Strains Tested	Pink/Blue-Turquoise-Green/Tan Colonies on ESBL Spectra	
		Negative	Positive
Acinetobacter baumannii	3	3	0
Acinetobacter lwoffii	1	1	0
Acinetobacter spp.	4	4	0
Aeromonas caviae	1	1	0
Aeromonas hydrophilia	4	4	0
Aeromonas veronii	1	1	0
Aspergillus brasiliensis	1	1	0
Candida albicans	1	1	0
Candida parapsilosis	1	1	0
Enterococcus faecalis	3 [1]	3	0
Enterococcus faecium	3	3	0
Lactobacillus acidophilus	2	2	0
Lactobacillus brevis	1	1	0
Lactobacillus casei	1	1	0
Lactobacillus delbruekii	1	1	0
Lactobacillus gasseri	1	1	0
Lactobacillus plantarum	1	1	0
Pediococcus pentosaceus	1	1	0
Penicillium chrysogenum	1	1	0
Pseudomonas aeruginosa	1	1	0
Saccharomyces cerevisiae	1	1	0
Staphylococcus aureus	5 [2]	5	0
Staphylococcus spp. (CoNS)	3	3	0
Stenotrophomonas maltophilia	4	4	0
Vibrio cholerae	1	1	0
Vibrio parahaemolyticus	1	1	0
Vibrio vulnificus	1	1	0

CoNS: Coagulase-Negative Staphylococci
[1] Including 1 strain of vancomycin-resistant *E. faecalis*
[2] All MRSA

Seventeen of the 18 non-ESBL producing strains of *Enterobacteriaceae* that produced pink/blue-turquoise-green/tan colonies on Spectra ESBL were non-susceptible to ceftazidime and cefotaxime (1 strain of *K. pneumoniae* was susceptible to both these antimicrobial agents). None of the non-*Enterobacteriaceae* tested produced colonies that could be mistaken for potentially ESBL producing species.

The potential for non-ESBL producing strains of *Enterobacteriaceae* that are non-susceptible to cephalosporins to be recovered on Spectra ESBL is noted as a Limitation in the Package Insert. The device labeling also stipulates that positive results on Spectra ESBL should not be reported and that subculture of presumptively positive colonies is required for organism identification, confirmatory testing for ESBL, susceptibility testing and epidemiological typing. The risks to patients from false positive results with Spectra ESBL are therefore mitigated and the results of the Analytical Specificity Study are acceptable.

e. *Assay cut-off:*

Not applicable.

g. *Assay Interference*

Interfering Microorganisms
A study was conducted to evaluate the ability of Spectra ESBL to recover target organisms from mixtures of ESBL-producing and non-ESBL producing species. Testing was performed by mixing target organisms at levels close the LOD of Spectra ESBL with non-target organisms at 10^6 to 10^9 CFU/mL in saline. The results of the Mixed Infection Study are summarized in **Table 5**.

Table 5. Summary of the results of the Mixed Infection Study

Non-target Organism		ESBL Producing Target Organism	
Species/Strain	**Observed Growth on Spectra ESBL**	**Species/Strain**	**Observed Growth on Spectra ESBL**
E. coli ATCC 25922	None	*K. pneumoniae* ATCC 700603	Yes
A. baumannii ATCC 19606	None	*K. pneumoniae* ATCC 700603	Yes
S. marcescens HIP 13096	Green colonies at $\geq 10^8$ CFU/mL	*E. coli* ATCC BAA-201	Yes
K. oxytoca HIP 15001	Green lawn at $\geq 10^6$ CFU/mL	*E. coli* ATCC BAA-201	*K. oxytoca* HIP 15001 $\leq 10^7$ CFU/mL: Yes *K. oxytoca* HIP 15001 $\geq 10^8$ CFU/mL: No
E. faecalis ATCC 51299 [1]	None [1]	*P. mirabilis* 511-116	Yes
L. brevis ATCC 8287	None [1]	*P. mirabilis* 511-116	Yes

[1] No growth evident due to swarming colony morphology of *P. mirabilis* target organism

The potential for high concentrations of non-ESBL producing organisms to produce colonies that are consistent in appearance with those of ESBL-producing strains and suppress growth of target organisms is noted as a Limitation in the device labeling. The Spectra ESBL Intended Use and result interpretation chart also stipulate that positive results should not be reported and that subculture of colonies is required for

organism identification and susceptibility testing, including ESBL confirmation.

Interfering Substances
A study was conducted to evaluate the performance of Spectra ESBL in the presence of endogenous and exogenous substances that may be present in stool and/or perirectal swab specimens. Twenty-one substances were tested in liquefied stool matrix in the presence of 5 representative ESBL-producing organisms (*E. coli* (2), *K. pneumoniae* (2) and *P. mirabilis* (1)). Semi-quantitative colony counts on Spectra ESBL were compared in the presence and absence of each potential interfering substance. The results are summarized in **Table 6** and are acceptable.

Table 6. Results of the Interfering Substances Study

Substance	Concentration at Which No Interference Was Observed
Barium sulfate	1.56 mg/mL [1]
Bisacodyl	50% (w/v) [2]
Blood	5.0% v/v
Fletcher's Castoria	10% v/v
Glycerin	50% (w/v) [2]
Imodium AD	10% v/v
Kaopectate	10% v/v
KY Jelly	5.0% w/v [1]
Metronidazole	1.56 mg/mL [1]
Miconazole	5.0% w/v [1]
Mucin	5% (w/v)
Mylanta ES	0.5% v/v [1]
Nanoxynol-9	50% w/v
Pepcid AC	0.5% v/v [1, 2]
Pepto-Bismol	10% v/v
Preparation H	5.0% w/v [1]
Preparation H Wipes	5.0% v/v [1]
Prilosec OTC	1.0% v/v [1, 2]
Tagamet HB 200	10% v/v [2]
Vancomycin	1.56 mg/mL [1]
Vaseline	5.0% w/v [1]

[1] Shown to be inhibitory at higher concentration
[2] One suppository or tablet dissolved in 10mL phosphate buffered saline (PBS), then added to stool

Charcoal Containing Transport Media
A study was conducted to evaluate the effect of charcoal-containing transport medium for perirectal swabs on the growth and colony color obtained on Spectra ESBL. Isolated colonies of 6 ESBL producing strains (*E. coli* (3), *K. pneumoniae* (2) and *P. mirabilis* (1)) were sampled using collection swabs that were then immersed in charcoal-containing transport medium prior to inoculating Spectra ESBL. The expected colony color was obtained with each ESBL-producing strain demonstrating

the compatibility of charcoal swabs with the culture medium. These results are acceptable.

2. Comparison studies:

   a. *Method comparison with predicate device:*

      Not applicable.

   b. *Matrix comparison:*

      Refer to **Section M(2)(g)**, above.

3. Clinical studies:

   a. *Clinical Sensitivity:*

      The performance of Spectra ESBL was evaluated in a prospective Clinical Study that was conducted at 3 sites using either fresh (unpreserved) stool or perirectal swab specimens that were collected from subjects at risk of colonization with ESBL-producing organisms. Two of the clinical sites tested stool specimens and a third site tested only perirectal swabs. To establish the presence or absence of ESBL positive organisms of the targeted species, reference culture was performed at each of the sites through inoculation of MacConkey agar plates with a 10µg cefpodoxime disk between the 1st and 2nd quadrants, followed by identification by standard methods and (as appropriate) ESBL confirmatory testing of colonies that grew in the zone of inhibition according to the procedure described in CLSI M100.[1] All specimens were cultured within 24 hours of collection.

      Spectra ESBL plates were read after 18 and 24 hours at 35±2°C and all colony types recovered were identified and, as appropriate, underwent confirmatory testing of the ESBL phenotype.

      Specimens from a total of 1194 subjects were enrolled in the study (748 fresh stool specimens and 446 perirectal swabs). Eighteen specimens were excluded from analysis due to protocol deviations: 15 (8 stool and 7 perirectal swab) because they were not plated within 24 hours of collection and 3 (all stool) because the isolates recovered were not available for identification and antimicrobial susceptibility testing. Isolates from a total 737 stool specimens and 439 perirectal swabs were therefore included in the analysis of performance.

      The results of the study are summarized in **Tables 7** to **11**. With perirectal swabs, the performance of Spectra ESBL was the same when plates were read after 18 and 24 hours, with sensitivity and specificity values of 99.1% (95% confidence interval:

[1] CLSI. Performance Standards for Antimicrobial Susceptibility Testing. 23rd ed. CLSI supplement M100. Wayne, PA: Clinical and Laboratory Standards Institute; 2013.

94.2-99.8%) and 89.7% (95% CI: 86.1-92.5%), respectively. For fresh stool specimens, the sensitivity and specificity values after 18 hours of incubation were 97.8% (95% CI: 92.5-99.4%) and 89.6% (95% CI: 87.1-91.7%), respectively, while after 24 hours the sensitivity was 98.9% (95% CI: 94.2-99.8%) and the specificity was 88.1% (95% CI: 85.5-90.3%). For both specimen types combined, the sensitivity and specificity were 98.5% (95% CI: 95.8-99.5%) and 89.6% (95% CI: 87.6-91.3%), respectively, after 18 hours and 99.0% (95% CI: 96.5-99.7%) and 88.7% (95% CI: 86.6-90.4%), respectively, after 24 hours.

Table 7. Performance of Spectra ESBL with perirectal swab specimens after 18 or 24 hour incubation vs reference culture with ESBL confirmation [1]

All Perirectal Swab Specimens Combined		**Reference Culture with ESBL Confirmation**		
		ESBL Positive EC, KP, KO or PM	**ESBL Negative or No Growth**	**Total**
Spectra ESBL	**Pink/Blue-Turquoise-Green/Tan Colonies**	110	36	**146**
	Other or No Growth	1	313	**314**
	Total	**111**	**349**	**460**
Sensitivity		110/111 = 99.1% (95% CI: 94.2-99.8%)		
Specificity		313/349 = 89.7% (95% CI: 86.1-92.5%)		
Positive Predictive Value		110/146 = 75.3%		
Negative Predictive Value		313/314 = 99.7%		

EC: *E. coli*; KP: *K. pneumoniae*; KO: *K. oxytoca*; PM: *P. mirabilis*

[1] For perirectal swabs, the same sensitivity and specificity were observed when plates were read at 18 and 24 hours

Table 8. Performance of Spectra ESBL with fresh stool specimens after 18 hour incubation vs reference culture with ESBL confirmation

All Fresh Stool Specimens Combined		**Reference Culture with ESBL Confirmation**		
		ESBL Positive EC, KP, KO or PM	**ESBL Negative or No Growth**	**Total**
Spectra ESBL 18 hours	**Pink/Blue-Turquoise-Green/Tan Colonies**	91	71	**162**
	Other or No Growth	2	612	**614**
	Total	**93**	**683**	**776**
Sensitivity		91/93 = 97.8% (95% CI: 92.5-99.4%)		
Specificity		612/683 = 89.6% (95% CI: 87.1-91.7%)		
Positive Predictive Value		91/162 = 56.2%		
Negative Predictive Value		612/614 = 99.7%		

EC: *E. coli*; KP: *K. pneumoniae*; KO: *K. oxytoca*; PM: *P. mirabilis*

Table 9. Performance of Spectra ESBL with fresh stool specimens after 24 hour incubation vs reference culture with ESBL confirmation

All Fresh Stool Specimens Combined		Reference Culture with ESBL Confirmation		
		ESBL Positive EC, KP, KO or PM	ESBL Negative or No Growth	Total
Spectra ESBL 24 hours	Pink/Blue-Turquoise-Green/Tan Colonies	93	81	**174**
	Other or No Growth	1	601	**602**
	Total	**94**	**682**	**776**
Sensitivity		93/94 = 98.9% (95% CI: 94.2-99.8%)		
Specificity		601/682 = 88.1% (95% CI: 85.5-90.3%)		
Positive Predictive Value		93/174 = 53.4%		
Negative Predictive Value		601/602 = 99.8%		

EC: *E. coli*; KP: *K. pneumoniae*; KO: *K. oxytoca*; PM: *P. mirabilis*

Table 10. Performance of Spectra ESBL with fresh stool and perirectal swab specimens combined vs reference culture with ESBL confirmation

18 hour incubation of Spectra ESBL		Reference Culture with ESBL Confirmation		
		ESBL Positive EC, KP, KO or PM	ESBL Negative or No Growth	Total
Spectra ESBL 18 hours	Pink/Blue-Turquoise-Green/Tan Colonies	201	107	**308**
	Other or No Growth	3	925	**928**
	Total	**204**	**1032**	**1236**
Sensitivity		201/204 = 98.5% (95% CI: 95.8-99.5%)		
Specificity		925/1032 = 89.6% (95% CI: 87.6-91.3%)		
Positive Predictive Value		201/308 = 65.3%		
Negative Predictive Value		925/928 = 99.7%		
24 hour incubation of Spectra ESBL		**Reference Culture with ESBL Confirmation**		
		ESBL Positive EC, KP, KO or PM	**ESBL Negative or No Growth**	**Total**
Spectra ESBL 24 hours	Pink/Blue-Turquoise-Green/Tan Colonies	203	117	**320**
	Other or No Growth	2	914	**916**
	Total	**205**	**1031**	**1236**
Sensitivity		203/205 = 99.0% (95% CI: 96.5-99.7%)		
Specificity		914/1031 = 88.7% (95% CI: 86.6-90.4%)		
Positive Predictive Value		203/320 = 63.4%		
Negative Predictive Value		914/916 = 99.8%		

EC: *E. coli*; KP: *K. pneumoniae*; KO: *K. oxytoca*; PM: *P. mirabilis*

In addition to comparing the results obtained with Spectra ESBL to those of the reference culture method, an additional analysis was performed to compare the colony colors obtained on Spectra ESBL with the organism identity and confirmed ESBL phenotype of the individual isolates recovered (**Table 11**). Colonies on Spectra ESBL were subcultured to sheep blood agar, identified biochemically and, as

appropriate, underwent confirmatory ESBL testing according to CLSI M100. Of 308 isolates on Spectra ESBL that exhibited pink/blue-turquoise-green/tan colonies after 18 hours, 203 were confirmed as ESBL producing strains of *E. coli*, *K. pneumoniae*, *K. oxytoca* or *P. mirabilis* (Positive Predictive Value [PPV] = 65.9%). After 24 hours, the PPV was 64.4% (206/320).

Because of the potential for false positive results with Spectra ESBL, the Intended Use and result interpretation table in the device labeling indicate that positive results should not be reported and that subculture is required for organism identification and ESBL confirmatory testing.

Table 11. Agreement between colony color obtained on Spectra ESBL and organism identity/confirmed ESBL phenotype at 18 and 24 hours

18 hours		**Organism Identity and Confirmed ESBL Phenotype**		
		ESBL Positive EC, KP, KO or PM	**ESBL Negative EC, KP, KO or PM or Other sp.**	**Total**
Spectra ESBL 18 hours	**Pink/Blue-Turquoise-Green/Tan Colonies**	203	105 [1]	**308**
	Other or No Growth	2	926	**928**
	Total	**205**	**1031**	**1236**
Positive Percent Agreement		203/205 = 99.0% (95% CI: 96.5-99.7%)		
Negative Percent Agreement		926/1031 = 89.8% (95% CI: 87.8-91.5%)		
Positive Predictive Value		203/308 = 65.9%		
Negative Predictive Value		926/928 = 99.8%		
24 hours		**Organism Identity and Confirmed ESBL Phenotype**		
		ESBL Positive EC, KP, KO or PM	**ESBL Negative EC, KP, KO or PM or Other sp.**	**Total**
Spectra ESBL 24 hours	**Pink/Blue-Turquoise-Green/Tan Colonies**	206	114 [2]	**320**
	Other or No Growth	0	916	**916**
	Total	**206**	**1030**	**1236**
Positive Percent Agreement		206/206 =100% (95% CI: 98.2-100%)		
Negative Percent Agreement		916/1030 = 88.9% (95% CI: 86.9-90.7%)		
Positive Predictive Value		206/320 = 64.4%		
Negative Predictive Value		916/916 = 100%		

[1] 104/105 *Enterobacteriaceae*: *Enterobacter* (38), *Citrobacter* (19), *E. coli* (20), *K. pneumoniae* (15), *K. oxytoc*a (7), Other (5)
[2] 113/114 *Enterobacteriaceae: Enterobacter* (38), *Citrobacter* (21), *E. coli* (21), *K. pneumoniae* (17), *K. oxytoc*a (8), Other (8)

b. Clinical specificity:

Refer to **Section M(3)(a)**, above.

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

Not Applicable

4. Clinical cut-off**:**

   Not Applicable

5. Expected values/Reference range:

   In the prospective Clinical Study described in **Section M(3)**, above, the prevalence of ESBL-producing *E.coli*, *K. pneumoniae*, *K. oxytoca* or *P. mirabilis* that was observed on Spectra ESBL after 18h incubation was 23.0% (270/1176) compared with 23.9% (281/1176) after incubation for 24h. As determined by the Reference Method after 18 and 24h incubation of the MacConkey plates, the prevalence was 15.0% (176/1176) and 15.3% (178/1176), respectively.

N. Proposed Labeling:

The labeling is sufficient and satisfies the requirements of 21 CFR 809.10.

O. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence determination.