

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

- A.** 510(k) Number:
K031965
- B.** Analyte:
Cryptosporidium antigens
- C.** Type of Test:
Chromatographic immunoassay
- D.** Applicant:
Remel Inc.
- E.** Proprietary and Established Names:
Xpect™ Cryptosporidium Lateral Flow Assay
- F.** Regulatory Information:
1. Regulation section:
21 CFR Part 866.3220 Entamoeba histolytica serological reagents
 2. Classification:
Class II
 3. Product Code:
MHJ – Cryptosporidium spp.
 4. Panel:
83 (Microbiology)
- G.** Intended Use:
1. Intended use(s):
Xpect™ Cryptosporidium kit is an in vitro qualitative immunoassay for the detection of Cryptosporidium antigens in preserved and unpreserved fecal specimens. This test is intended as an aid in the laboratory diagnosis of suspected Cryptosporidium infections.
 2. Indication(s) for use:
Xpect™ Cryptosporidium kit is an in vitro qualitative immunoassay for the detection of Cryptosporidium antigens in preserved and unpreserved fecal specimens. This test is intended as an aid in the laboratory diagnosis of suspected Cryptosporidium infections.
 3. Special condition for use statement(s):
Not applicable
 4. Special instrument Requirements:
Not applicable
- H.** Device Description:
The kit contains 20 test devices consisting of a membrane striped with rabbit anti-Cryptosporidium, and goat anti-mouse IgG. A mixture of specimen, buffered diluent, and conjugate containing colored micro-particles linked to murine monoclonal antibody specific for Cryptosporidium, is dispensed into the sample well of the device. The test sample wicks along a membrane containing capture antibody stripes. The Cryptosporidium immune complex, if present, reacts with

anti-Cryptosporidium antibody at the test line. Conjugate not bound at the test line is later captured at the control line containing anti-mouse antibody. A red line of any intensity will appear at the Cryptosporidium test position if Cryptosporidium antigen is present. A line in the Control position indicates that the test is working properly. The device also contains a procedure card; disposable transfer pipettes, dilution tubes, and instructions for use.

I. Substantial Equivalence Information:

1. Predicate device name(s):
BD ColorPAC Giardia/Crypto Rapid Assay
2. Predicate K number(s):
K 983399
3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Intended Use	Detection of Cryptosporidium antigens in fecal specimens	Detection of Giardia and Cryptosporidium antigens in fecal specimens
Technology	Qualitative immunochromatographic assay	Qualitative immunochromatographic assay
Antibodies:conjugate	Monoclonal anti-Cryptosporidium	Rabbit anti-Giardia, monoclonal anti-Giardia and Cryptosporidium
Specimen type	Human stool preserved in 10% formalin, SAF, MIF or Cary Blair	Human stool preserved in 10% formalin, SAF, MIF, or Cary Blair
Differences		
Item	Device	Predicate
Capture antibodies or molecules	Rabbit anti-Cryptosporidium, goat anti-mouse IgG	Mouse anti-Cryptosporidium, goat anti-mouse IgG, Avidin derivative
Material: membrane	Mylar-backed nitrocellulose	Nitrocellulose
Material: conjugate	Anti-mouse and anti-Cryptosporidium colored polystyrene microparticles diluted in buffer	Colloidal dye labeled monoclonal antibodies to Giardia and Cryptosporidium
Sample volume	100µl	50µl

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

Not applicable

K. Test Principle:

The Xpect™ Cryptosporidium Lateral Flow Assay is a chromatographic immunoassay that detects the presence of Cryptosporidium antigens. The test utilizes sample wicking to capture Cryptosporidium antigens on discrete test lines containing antigen-specific antibodies for each organism. A specimen is added to a dilution tube containing a buffered solution. A conjugate containing colored micro-particles linked to monoclonal antibodies specific for Cryptosporidium is added. The mixture is dispensed into the sample well of the device and wicks across a membrane containing capture antibody strips. The Cryptosporidium immune complexes, if present, react with anti-Cryptosporidium antibody at the test line. Conjugates not bound at the test lines are later captured at the control line containing anti-mouse antibody. A pink line will appear at the Cryptosporidium test position if Cryptosporidium antigen is present. A line in the Control position indicates that the test is working properly.

L. Performance Characteristics (if/when applicable):1. Analytical performance:

a. Precision/Reproducibility:

Reproducibility testing was conducted at seven sites, including one in-house site, on three separate days with ten blinded samples of varying activity. All samples tested for Cryptosporidium produced the expected result.

b. Linearity/assay reportable range:

Not applicable

c. Traceability (controls, calibrators, or method):

Not applicable

d. Detection limit:

For Cryptosporidium, detection limit was 6 organisms per 0.1 ml. of specimen

e. Analytical specificity:

Cross-reactivity:

No cross-reactivity was observed using samples containing the following organisms: *Ascaris lumbricoides*, *Blastocystis hominis*, *Campylobacter coli*, *Campylobacter jejuni*, *Candida albicans*, *Chilomastix mesnili*, *Cyclospora cayetanensis*, *Dientamoeba fragilis*, *Endolimax nana*, *Entamoeba coli*, *Entamoeba hartmanni*, *Entamoeba histolytica*, *Enterobius vermicularis*, *Escherichia coli*, hookworm, *Hymenolepis nana*, *Iodamoeba bütschlii*, *Isospora sp.*, *Microsporidia*, *Rotavirus*, *Salmonella choleraesuis* subsp. *choleraesuis* serotype *Typhimurium*, *Shigella dysenteriae*, *Shigella flexneri*, *Shigella sonnei*, *Strongyloides stercoralis*, *Taenia sp.*, and *Trichuris trichiura*. Cross-reactivity to *Astrovirus* and *Caliciviruses* has not been established.

Interfering Substances:

Prior to testing, positive and negative samples were spiked (20% v/v) with blood, mucin, fecal fat or the following over-the-counter anti-diarrheal products: *Pepto-Bismol*®, *Imodium*® A-D, and *Kaopectate*® (active ingredients: bismuth

subsalsicylate, loperamide HCl, and attapugite respectively). Testing indicated that none of these substances interfered with the expected result except for Imodium® A-D. Imodium® A-D at 20% (v/v) in stool interfered with the detection of low levels of Cryptosporidium antigen.

f. Assay cut-off:

The assay can detect 6 Cryptosporidium organisms per 0.1 ml of specimen. Clinically relevant detection limits were determined using true clinical specimens diluted to an end point titration with the Xpect Cryptosporidium test. Sequential serial dilutions were tested until an endpoint dilution was reached. The endpoint dilution was defined as one dilution above where the sample became negative. The quantity of organisms detected at the titration endpoint in each specimen was calculated from the numbers seen microscopically in a 10 µl sample using DFA.

2. Comparison studies:

a. Method comparison with predicate device:

Percent Agreement: Xpect™ & Predicate Device vs. Microscopy:

The Xpect™ Cryptosporidium and a commercially available lateral flow test (the “Predicate Device”) were compared side-by-side to microscopy. The Percent Agreement of each assay versus microscopy was as follows:

Cryptosporidium		Microscopy		
		+	-	
<u>Xpect™</u>	+	28	6	Agreement
	-	2	110	
Total		30	116	94.5% (138/146)

<u>Predicate</u>		+	-	Agreement
		29	21	
1	95			
Total		30	116	84.9% (124/146)

b. Matrix comparison:

Not applicable

3. Clinical studies:

a. Clinical sensitivity:

Sensitivity and Specificity Compared to Microscopy:

The performance of the Xpect™ Cryptosporidium was evaluated at six geographically diverse laboratories. The overall sensitivity and specificity of the test was compared to microscopy using 578 specimens. Samples tested were

samples routinely submitted for microscopic exam. Performance relative to patients' clinical status has not been established. Data is listed below.

Cryptosporidium		Microscopy	
		+	-
Xpect™	+	108	8
	-	4	458
Total		112	466

Sensitivity: 96.4% (108/112); 95% CI = 91.2-98.6%

Specificity: 98.3% (458/466); 95% CI = 96.6-99.1%

- b. Clinical specificity:
Refer to (a) above
- c. Other clinical supportive data (when a and b are not applicable):
Not applicable
4. Clinical cut-off:
See assay cut off above
5. Expected values/Reference range:
Expected values were established from literature. Prevalence rates for Cryptosporidium range from 1-3% in North America and Europe to 7-9% in less developed countries. Rates tend to be higher in children under 2 years and in persons with weakened immune systems.

M. Conclusion:

In clinical settings, the Xpect™ Cryptosporidium Lateral Flow Assay is substantially equivalent in performance to the predicate device and to microscopic examination for the identification of Cryptosporidium in fecal specimens.