

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

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

K103673

**B. Purpose for Submission:**

To obtain a substantial equivalence determination for the detection of *Giardia* and *Cryptosporidium* antigens in human stool

**C. Measurand:**

*Giardia* and *Cryptosporidium* antigens

**D. Type of Test:**

Qualitative membrane enzyme immunoassay

**E. Applicant:**

TechLab<sup>®</sup> Inc.

**F. Proprietary and Established Names:**

The *GIARDIA/CRYPTOSPORIDIUM* QUIK CHEK<sup>™</sup> test

**G. Regulatory Information:**

1. Regulation section:

21 CFR Part 866.3220 *Entamoeba histolytica* Serological Reagents

2. Classification:

II

3. Product code:

MHJ - *Cryptosporidium* spp.

MHI – *Giardia* spp.

4. Panel:

83, Microbiology

**H. Intended Use:**

1. Intended use(s):

The GIARDIA/CRYPTOSPORIDIUM QUIK CHEK test is a rapid membrane enzyme immunoassay for the simultaneous qualitative detection and differentiation of *Giardia* cyst antigen and *Cryptosporidium* oocyst antigen in a single test device. It is intended for use with human fecal specimens from patients with gastrointestinal symptoms to aid in the diagnosis of *Giardia* and/or *Cryptosporidium* gastrointestinal infection. As with other *Giardia* and/or *Cryptosporidium* tests, results should be considered in conjunction with the patient history.

FOR IN VITRO DIAGNOSTIC USE.

2. Indication(s) for use:

The GIARDIA/CRYPTOSPORIDIUM QUIK CHEK test is a rapid membrane enzyme immunoassay for the simultaneous qualitative detection and differentiation of *Giardia* cyst antigen and *Cryptosporidium* oocyst antigen in a single test device. It is intended for use with human fecal specimens from patients with gastrointestinal symptoms to aid in the diagnosis of *Giardia* and/or *Cryptosporidium* gastrointestinal infection. As with other *Giardia* and/or *Cryptosporidium* tests, results should be considered in conjunction with the patient history.

3. Special conditions for use statement(s):

For prescription use only

4. Special instrument requirements:

None

**I. Device Description:**

The GIARDIA/CRYPTOSPORIDIUM QUIK CHEK™ test device contains 25 pouches, each containing a membrane filter pad. Each device has a *Reaction Window* with three vertical lines of immobilized antibodies. The *Giardia* test line (“Giar”) contains mouse monoclonal antibodies against *Giardia*. The *Crypto* test line (“Cryp”) contains mouse monoclonal antibodies against *Cryptosporidium*. The control line (“C”) is a dotted line that contains anti-horseradish peroxidase (HRP) antibodies. The

*Conjugate* consists of polyclonal antibodies coupled to horseradish peroxidase. The antigen-antibody-conjugate complexes migrate through a filter pad to a membrane where they are captured by the immobilized *Giardia* and/or *Cryptosporidium*-specific antibodies in the test lines. After addition of substrate and proper incubation, the reaction is examined visually for the appearance of a vertical blue line on either side of the Reaction Window. A blue line indicates a positive test. A positive “control” reaction, indicated by a vertical dotted blue line under the “C” portion of the Reaction Window, confirms that the test is working properly and the results are valid.

**J. Substantial Equivalence Information:**

1. Predicate device name(s):

GIARDIA II  
CRYPTOSPORIDIUM II

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

K033274  
K052932

3. Comparison with predicate:

<b>Similarities</b>			
Item	Device	Predicate 1	Predicate 2
Intended Use	Detection of <i>Giardia</i> cyst and <i>Cryptosporidium</i> oocyst antigen	Detection of <i>Giardia</i> cyst antigen	Detection of <i>Cryptosporidium</i> oocyst antigen
Technology	Enzyme Immunoassay	Enzyme Immunoassay	Enzyme Immunoassay
Antibody Format	Monoclonal/Polyclonal	Monoclonal/Polyclonal	Monoclonal/Polyclonal
Specimen Types	Human Fecal Specimens- fresh, frozen, or preserved in 10% Formalin or SAF	Human Fecal Specimens- fresh, frozen, or preserved in 10% Formalin or SAF	Human Fecal Specimens- fresh, frozen, or preserved in 10% Formalin or SAF

Differences			
Item	Device	Predicate 1	Predicate 2
Intended Use	Detection and differentiation of <i>Giardia</i> cyst antigen and <i>Cryptosporidium</i> oocyst antigen	Detection <i>Giardia</i> cyst antigen only	Detection <i>Cryptosporidium</i> oocyst antigen only
Technology	Membrane Enzyme Immunoassay	Enzyme Immunoassay – Microassay Plate ELISA	Enzyme Immunoassay – Microassay Plate ELISA
Amount of Specimen required	25 µL – fresh 100 µL - preserved	100 µL – fresh or preserved	100 µL – fresh or preserved
Incubation time	15 minutes	1 hour	1 hour
Time to Result	30 minutes	1 hour 45 minutes	1 hour 45 minutes

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

Not applicable

**L. Test Principle:**

The *GIARDIA/CRYPTOSPORIDIUM QUIK CHEK™* test is a membrane enzyme immunoassay that uses monoclonal and polyclonal antibodies to cell-surface antigens of the organisms. The polyclonal antibodies are coupled to horseradish peroxidase which are contained in the diluent. Specific mouse monoclonal antibodies against *Giardia* and *Cryptosporidium* antigens are bound on different location of the membrane. A fecal specimen is added to the diluent and transferred to the sample well and incubated. During the incubation, cyst and/or oocyst antigens in the sample bind the antibody-peroxidase conjugates. The antigen-antibody-conjugate complexes migrate through a filter pad to a membrane where they are captured by the immobilized *Giardia* and/or *Cryptosporidium*-specific antibodies in the test lines.

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

1. Analytical performance:

a. *Precision/Reproducibility:*

A total of 22 fecal specimens were pre-characterized by commercially available predicate devices. The samples included six *Giardia*-positive specimens (three mid-range positives), six *Cryptosporidium*-positive

specimens (three low positives), and four *Giardia/Cryptosporidium*-positive specimens (two of which were low *Giardia*-positives and two of which were low *Cryptosporidium*-positives), and six specimens negative for both parasites. All specimens were coded to prevent their identification during testing. Testing was performed at three sites. The samples were tested, twice a day over a 5-day period by multiple technicians at each site using two different kit lots. A positive and negative control was run with each panel of the masked samples. The results were consistent among the different locations, and exhibited a correlation of 100%.

b. *Linearity/assay reportable range:*

Not applicable

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

Not applicable.

d. *Detection limit:*

The analytical sensitivity of the *GIARDIA/CRYPTOSPORIDIUM QUIK CHEK* test was determined by spiking various dilutions of purified *Giardia* cysts or *Cryptosporidium* oocysts into negative human stool. The cyst/oocyst preparations were quantified using the reference assay of IFA microscopy. Test results determined the limit of detection to be 6000 *Giardia* cysts per mL (133 cysts/test) and 6000 *Cryptosporidium* oocysts/mL (133 cysts/test).

e. *Analytical specificity:*

Cross- Reactivity Study

No cross reactions were seen with the following organisms:

*Aeromonas hydrophila*, *Bacillus cereus*, *Bacillus subtilis*, *Bacteroides fragilis*, *Campylobacter coli*, *Campylobacter fetus*, *Campylobacter jejuni*, *Candida albicans*, *Clostridium bifermentans*, *Clostridium difficile* (strain 630), *Enterococcus faecalis*, *Escherichia coli*, *Escherichia coli* 0157:H7, *Escherichia coli* ETEC (enterotoxigenic), *Escherichia coli* EPEC (enteropathogenic), *Escherichia coli* EIEC (enteroinvasive), *Klebsiella pneumoniae*, *Salmonella typhimurium*, *Shigella dysenteriae*, *Shigella flexneri*, *Shigella sonnei*, *Staphylococcus aureus*, *Staphylococcus aureus* (Cowan's), *Staphylococcus epidermidis*, *Vibrio parahaemolyticus*, *Yersinia enterocolitica*

Human Adenovirus 1 and 3, Adenovirus, Type 2, 5, 40 and 41, Human Coxsackievirus B2, B3, and B4, Coxsackievirus B5, Human Coronavirus, Echovirus 11, 18, 33, Human Echovirus 9, Human paraechovirus 1 (Echovirus 22), Enterovirus 68, 69, Human Enterovirus 70, 71, Human rotavirus

Additionally, the GIARDIA/CRYPTOSPORIDIUM QUIK CHEK™ test was tested using fecal specimens documented to be positive for other parasites by microscopy; *Ascaris lumbricoides* eggs, *Dientamoeba fragilis*, *Entamoeba hartmanni*, *Blastocystis hominis*, *Diphyllobothrium latum* eggs, *Entamoeba histolytica/E. dispar*, *Chilomastix mesnili*, *Endolimax nana*, Hookworm eggs, *Cyclospora cayetanensis*, *Entamoeba coli*, *Iodamoeba butschlii*, and *Trichuris trichiura* eggs. No cross-reactivity was seen with the following organisms for either the *Giardia*-portion or the *Cryptosporidium*-portion of the test.

Cross-reactivity to Astrovirus and Caliciviruses has not been evaluated.

The results obtained from the cross reactive study is acceptable.

#### Interference Study

The following substances had no effect on positive or negative test results analyzed at the concentrations indicated: Hog gastric mucin (3.5% w/v), Human blood (40% v/v), Barium sulfate (5% w/v), Imodium (5% v/v), Kaopectate (5% v/v), Pepto-Bismol (5% v/v), Steric/Palmitic Acid (40% w/v), Metronidazole (0.25% w/v), Vancomycin (0.25% w/v).

The results obtained from the interference study is acceptable.

#### f. Assay cut-off:

Not Applicable

## 2. Comparison studies:

### a. *Method comparison with predicate device:*

The performance of the GIARDIA/CRYPTOSPORIDIUM QUIK CHEK test was evaluated at three geographically diverse sites. Two sites compared the performance to commercially available ELISA (separate devices for *Giardia* and *Cryptosporidium*). A total of 849 specimens were evaluated and included 349 fresh, 322 frozen, 36 preserved (formalin) and 142 preserved (SAF).

Two hundred and fifteen (215) specimens were positive for *Giardia* by ELISA. The GIARDIA/CRYPTOSPORIDIUM QUIK CHEK test exhibited a 99.1% agreement for *Giardia* positive specimens, a 99.7% agreement for *Giardia* negative specimens, with an overall agreement of 99.5%.

One hundred thirty one (131) specimens were positive for *Cryptosporidium* by ELISA. The GIARDIA/CRYPTOSPORIDIUM QUIK CHEK test exhibited a 99.2% agreement for *Cryptosporidium* positive specimens, 99.6% agreement for *Cryptosporidium* negative specimens, and an overall

agreement of 99.5%.

The results obtained from the method comparison study are acceptable.

b. *Matrix comparison:*

Not Applicable

3. Clinical studies:

a. *Clinical Sensitivity:*

The performance of the GIARDIA/CRYPTOSPORIDIUM QUIK CHEK test was evaluated at three geographically diverse sites. Two sites compared performance to the reference microscopy/immunofluorescence (IFA). A total of 791 were evaluated and included 220 fresh, 140 frozen, 216 preserved (formalin) and 215 preserved (SAF).

One hundred eighty three (183) specimens were positive for *Giardia* by IFA. The GIARDIA/CRYPTOSPORIDIUM QUIK CHEK test exhibited a sensitivity of 98.9%, a specificity of 100%, and an overall correlation of 99.7% with microscopy.

One hundred forty (140) specimens were positive for *Cryptosporidium* by IFA. The GIARDIA/CRYPTOSPORIDIUM QUIK CHEK test exhibited a sensitivity of 100%, a specificity of 99.8%, and an overall correlation of 99.9% with Microscopy.

The results obtained from the clinical study appear to be acceptable.

**Combined Results - Clinical Performance Comparing the *Giardia* Line of the GIARDIA/CRYPTOSPORIDIUM QUIK CHEK Test to Microscopy – IFA**

N = 791	Microscopy - IFA <i>Giardia</i> positive	Microscopy - IFA <i>Giardia</i> negative
GIARDIA/CRYPTOSPORIDIUM QUIK CHEK <i>Giardia</i> Line Positive	181	0
GIARDIA/CRYPTOSPORIDIUM QUIK CHEK <i>Giardia</i> Line Negative	2	608

		95% Confidence Limits
Sensitivity	98.9%	95.7 – 99.8%
Specificity	100%	99.2 – 100%
Correlation	99.7%	99.7 – 99.7%

**Combined Results - Clinical Performance Comparing the  
Cryptosporidium Line of the GIARDIA/CRYPTOSPORIDIUM QUIK  
CHEK Test to Microscopy – IFA**

N = 791	Microscopy - IFA <i>Crypto.</i> positive	Microscopy - IFA <i>Crypto.</i> negative
<i>GIARDIA/CRYPTOSPORIDIUM QUIK CHEK Cryptosporidium Line Positive</i>	140	1
<i>GIARDIA/CRYPTOSPORIDIUM QUIK CHEK Cryptosporidium Line Negative</i>	0	650

		95% Confidence Limits
<b>Sensitivity</b>	100%	96.7 – 100%
<b>Specificity</b>	99.8%	99.0 – 100%
<b>Correlation</b>	99.9%	100 – 100%

b. *Clinical specificity:*

See section M.3.a

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:

Normal healthy individuals should not be infected with *Giardia* or *Cryptosporidium* and should test negative in the *GIARDIA/CRYPTOSPORIDIUM QUIK CHEK* test. A positive test result in the *GIARDIA/CRYPTOSPORIDIUM QUIK CHEK* test indicates that the person is shedding detectable amounts of *Giardia* and/or *Cryptosporidium* antigen. The incidence of *Giardia* and *Cryptosporidium* infection varies significantly between populations and geographic regions. Children in daycare settings have exhibited higher rates of infection with *Giardia* than the normal population. In addition, homosexual men have shown higher rates of infection. In general, laboratory-confirmed incidence of cryptosporidiosis in developed countries ranges from 1 to 2% overall with a higher incidence in children.

Prospective studies to evaluate the *GIARDIA/CRYPTOSPORIDIUM QUIK CHEK* test were conducted at an Asian hospital specializing in diarrheal diseases. The studies included 129 freshly collected fecal specimens. A positive test result in the

*GIARDIA/CRYPTOSPORIDIUM QUIK CHEK* test was obtained in 22.5% for *Giardia* and in 5.4% for *Cryptosporidium* antigen.

**N. Proposed Labeling:**

The labeling is sufficient and 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.