Rapid Response™

***Entamoeba histolytica* Test Cassette** (Feces)

REF ENT-9C10 Product Insert

A rapid, one step test for the qualitative detection of *Entamoeba histolytica* antigens in human feces.

For professional *in vitro* diagnostic use only.

Intended Use

The Rapid Response™ *Entamoeba histolytica* Test Cassette (Feces) is a rapid chromatographic immunoassay for the qualitative detection of *Entamoeba histolytica* antigens in human feces.

Introduction

Entamoeba histolytica is an anaerobicparasiticamoebozoan, part of the genus Entamoeba.[1] Predominantly infecting humans and other primates causing amoebiasis, *E. histolytica* is estimated to infect about 50 million people worldwide. Previously, it was thought that 10% of the world population was infected, but these figures predate the recognition that at least 90% of these infections were due to a second species, *E. dispar*.[2] Mammals such as dogs and cats can become infected transiently but are not thought to contribute significantly to transmission. *E. histolytica*, as its name suggests (histolytic= tissue destroying), is pathogenic; infection can be asymptomatic or can lead to amoebic dysenteryor amoebicliver abscess.[1, 3]

Principle

The Rapid Response™ *Entamoeba histolytica* Test Cassette (Feces) is a qualitative, lateral flow immunoassay for the detection of Entamoeba histolytica antigens in human feces. The membrane is precoated with anti-*Entamoeba histolytica* antibody on the test line region of the test. During testing, E. histolytica antigens, if present in the specimen react with *Entamoeba histolytica* antibodies conjugated colored particles. The antigen-conjugate complex migrates upward on the membrane chromatographically by capillary action to react with anti-*Entamoeba histolytica* antibodies on the membrane and generate a colored line. The presence of this colored line in the test line region indicates a positive result, while its absence indicates a negative result. To serve as a procedural control, a colored line will always appear in the control line region, indicating that the proper volume of specimen has been added and membrane wicking has occurred.

Reagents

The test contains anti-*Entamoeba histolytica* antibody conjugated colored particles and anti-*Entamoeba histolytica* antibodies coated on the membrane.

Precautions

- For professional *in vitro* diagnostic use only. Do not use after expiration date.
- The test should remain in the sealed pouch until use.
- Do not eat, drink or smoke in the area where the specimens or kits are handled.
- Handle all specimens as if they contain infectious agents. Observe established precautions against microbiological hazards throughout all procedures and follow the standard procedures for proper disposal of specimens.
- Wear protective clothing such as laboratory coats, disposable gloves, and eye protection when specimens are assayed.
- The used test should be discarded according to local regulations.
- Humidity and temperature can adversely affect results.

Materials

Materials provided

- Test cassettes
- Specimen collection tubes with extraction buffer
- Product insert
- Droppers

Materials required but not provided

- Specimen collection containers
- Timer

Storage and Stability

- The kit can be stored at room temperature or refrigerated 36-86°F (2-30°C). The test cassette is stable through the expiration date printed on the sealed pouch.
- The test cassette must remain in the sealed pouch until use.
- **Do not freeze.**
- Do not use beyond the expiration date.

Collection and Storage of Specimens

1. The feces specimen must be collected in clean, dry, waterproof container containing no detergents, preservatives or transport media.
2. Bring the necessary reagents to room temperature before use.

Test Procedure

Allow the test, specimen, buffer and/or controls to reach room temperature (59-86°F; 15-30°C) prior to testing.

To Collect Fecal Specimens:

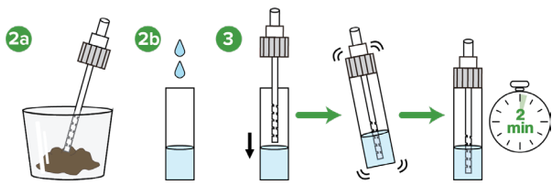
1. Collect sufficient quantity of feces (1-2 mL or 1-2 g) in a clean, dry specimen collection container to obtain maximum antigens (if present). Best results will be obtained if the assay is performed within 6 hours after collection. Specimen collected may be stored for 3 days at 36-46°F (2-8°C) if not tested within 6 hours. For long term storage, specimens should be kept below -4°F (-20°C).

To Process Fecal Specimens:

2a. **For Solid Specimens:**
Unscrew the cap of the specimen collection tube, then insert the applicator stick into the fecal specimen at least 3 different sites to collect approximately 50 mg of feces (equivalent to 1/4 of a pea). Do not scoop the fecal specimen.

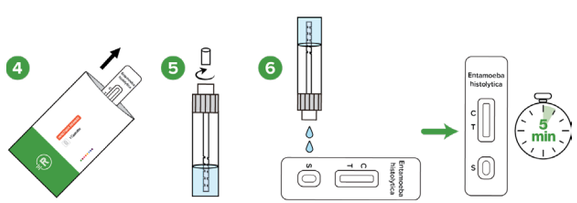
2b. **For Liquid Specimens:**
Hold the dropper vertically, aspirate fecal specimens, and then transfer 2 drops (approximately 80µL) into the specimen collection tube containing the extraction buffer.

3. Tighten the cap onto the specimen collection tube, then shake the specimen collection tube vigorously to mix the specimen and the extraction buffer. Leave the tube alone for 2 minutes.

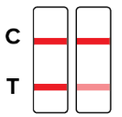


Test Sample Collection:

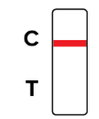
4. Bring the pouch to room temperature before opening it. Remove the test cassette from the foil pouch and use it within one hour. Best results will be obtained if the test is performed immediately after opening the foil pouch.
5. Hold the specimen collection tube upright and open the cap onto the specimen collection tube. Invert the specimen collection tube and transfer 2 full drops of the extracted specimen (approximately 80µL) to the specimen well (S) of the test cassette, then start the timer. Avoid trapping air bubbles in the specimen well (S).
6. Read results 5 minutes after dispensing the specimen. Do not read results after 10 minutes.
7. **Note:** If the specimen does not migrate, centrifuge the extracted specimens contained in the extraction buffer vial. Collect 80µL of supernatant, dispense into the specimen well (S) of a new test cassette and start afresh following the instructions mentioned above.



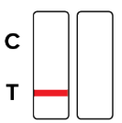
Results Interpretation



POSITIVE:* Two colored lines appear. One colored line should be in the control line region (C) and another apparent colored line should be in the test line region (T).
***NOTE:** The intensity of the color in the test line region (T) will vary depending on the concentration of *Entamoeba histolytica* antigen present in the specimen. Therefore, any shade of color in the test line region (T) should be considered positive.



NEGATIVE: One colored line appears in the control line region (C). No line appears in the test line region (T).



INVALID: Control line fails to appear. Insufficient specimen volume or incorrect procedural techniques are the most likely reasons for control line failure. Review the procedure and repeat the test with a new cassette. If the problem persists, discontinue using the test kit immediately and contact your local distributor.

Quality Control

- Internal procedural controls are included in the test. A colored line appearing in the control region (C) is an internal valid procedural control. It confirms sufficient specimen volume and correct procedural technique.
- Control standards are not supplied with this kit; however, it is recommended that positive and negative controls be tested as a good laboratory practice to confirm the test procedure and to verify proper test performance.

Limitations

1. The Rapid Response™ *Entamoeba histolytica* Test Cassette (Feces) is for *in vitro* diagnostic use only.
2. The Rapid Response™ *Entamoeba histolytica* Test Cassette (Feces) will only indicate the presence of *Entamoeba histolytica* antigen in the feces specimen, the detail concentration of *Entamoeba histolytica* antigen was not confirmed with the rapid test.
3. As with all diagnostic tests, all results must be considered with other clinical information available to the physician.
4. Other clinically available tests are required if questionable results are obtained.

Performance Characteristics

Clinical Sensitivity, Specificity and Accuracy

Document Number: 146666401
Revision Date: 2023-05-04

The performance of the Rapid Response™ *Entamoeba histolytica* Test Cassette (Feces) has been evaluated with 142 clinical specimens collected from the patient symptomatic and asymptomatic in comparison with other rapid test method. The results show that the relative sensitivity of the Rapid Response™ *Entamoeba histolytica* Test Cassette (Feces) is 95.7% and the relative specificity is 99.2%.

Method		Other Rapid Test		Total Results
Rapid Response™ *Entamoeba histolytica* Test Cassette (Feces)	Results	Positive	Negative	
	Positive	22	1	23
	Negative	1	118	119
Total Results		23	119	142

Relative Sensitivity: 95.7% (95%CI*: 78.1%~99.9%);
Relative Specificity: 99.2% (95%CI*: 95.4%~99.9%);
Overall Accuracy: 98.6% (95%CI*: 95.0%~99.8%).
*Confidence Intervals

Precision

Intra-Assay

Within-run precision has been determined by using 3 replicates of these specimens: negative, low positive, middle positive and high positive specimens. The specimens were correctly identified >99% of the time.

Inter-Assay

Between-run precision has been determined by 3 independent assays on the same specimens: negative, low positive, middle positive and high positive specimens. Three different lots of the Rapid Response™ *Entamoeba histolytica* Test Cassette (Feces) have been tested using these specimens. The specimens were correctly identified >99% of the time.

Cross Reactivity

An evaluation was performed to determine the cross reactivity of Rapid Response™ *Entamoeba histolytica* Test Cassette (Feces). No cross reactivity against gastrointestinal pathogens occasionally present as following:

Campylobacter coli	*Campylobacter jejuni*
E.coli O157:H7	*H.pylori*
Salmonella enteritidis	*Salmonella paratyphi*
Salmonella typhi	*Salmonella typhimurium*
Colstridiumdifficile	*Staphylococcus aureus*
Yersinia enterocolitica ersinia	*Listeria monocytogene*

Bibliography

1. Ryan KJ, Ray CG, eds. (2004).Sherris Medical Microbiology(4th ed.). McGraw Hill. pp.733–8.
2. Weekly Epidemiological Record.72(14): 97–9. April 1997.
3. Nespola, Benoît; Betz, Valérie; Brunet, Julie; Gagnard, Jean-Charles; Krummel, Yves; Hansmann, Yves; Hannedouche, Thierry; Christmann, Daniel; Pfaff, Alexander W.; Filisetti, Denis; Pesson, Bernard; Abou-Bacar, Ahmed; Candolfi, Ermanno (2015)."First case of amebic liver abscess 22 years after the first occurrence"

Glossary of Symbols

Consult instructions for use	Test per Kit	EC REP Authorized Representative in EU
Store between 36°F to 86°F	Use by	Do Not Reuse
LOT Lot Number	IVD For *in vitro* diagnostic use only	REF Catalogue #
Caution	Do not use if package is damaged	Manufacturer

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CE

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