

# VITASSAY

## Crypto+Giardia

Rapid test for the simultaneous qualitative detection of Cryptosporidium and Giardia in human stool samples.

IUE-7455035 Ed00 May 2016



For professional *in vitro* diagnostic use only.

### INTENDED USE

**Vitassay Crypto+Giardia** is a rapid, immunochromatographic, one step assay for the simultaneous qualitative detection of Cryptosporidium and Giardia in human stool samples.

Simple, non-invasive and highly sensitivity immunoassay to make a presumptive diagnosis of cryptosporidiosis and/or giardiasis.

### INTRODUCTION

Diarrheal diseases are extremely common in the developed and developing worlds and are major causes of morbidity and mortality, affecting millions of individuals each year. The etiologies of diarrhea include viruses, bacteria and parasites of which Entamoeba histolytica, Giardia lamblia and Cryptosporidium parvum are considered to be the most important.

Giardia lamblia and Cryptosporidium spp. are common protozoan parasites in humans, causing intestinal infections with watery diarrhea, abdominal pain and malabsorption that may last for weeks or months.

### PRINCIPLE

**Vitassay Crypto+Giardia** is a qualitative immunochromatographic assay for the detection of Cryptosporidium and Giardia in human stool samples.

**Strip A:** The test line zone of the nitrocellulose membrane is pre-coated with monoclonal antibodies against Crypto.

**Strip B:** The test line zone of the nitrocellulose membrane is pre-coated with monoclonal antibodies against Giardia.

During the process, the sample reacts with the antibodies against Crypto (strip A) and/or Giardia (strip B), forming conjugates. The mixture moves upward on the membrane by capillary action. If the sample is Crypto positive, antibodies present on the membrane (test line) capture the conjugate complex and a red line will be visible in the strip A, and if the sample is Giardia positive, antibodies present on the membrane (test line) capture the conjugate complex and a red line will be visible in strip B. Although the sample is positive or negative, the mixture continues to move across the membranes and the green control line always appears (for both strips).

The presence of these green lines (in the control zone (C)) indicates that sufficient volume is added; proper flow is obtained and serves as an internal control for the reagents.

### PRECAUTIONS

- For professional *in vitro* use only.
- Do not use after expiration date.
- Do not use the test if its pouch is damaged.
- Specimens should be considered as potentially hazardous and handle in the same manner as an infectious agent. A new test must be used for each sample to avoid contaminations errors.
- Tests should be discarded in a proper biohazard container after testing.
- Reagents contain preservatives. Avoid any contact with the skin or mucous membrane. Consult safety data sheet, available on request.
- Components provided in the kit are approved for use with the **Vitassay Crypto+Giardia**. Do not use any other commercial kit component.
- Follow Good Laboratory Practices, wear protective clothing, use disposal gloves, goggles and mask. Do not eat, drink or smoke in the working area.

### STORAGE AND STABILITY

Store as packaged in the sealed pouch either at refrigerated or room temperature (2-30°C/35.6-86°F).

The test is stable until the expiration date printed on the sealed pouch.

The test must remain in the sealed pouch until use.

Do not freeze.

### MATERIALS

MATERIAL PROVIDED	MATERIAL REQUIRED BUT NOT PROVIDED
<ul style="list-style-type: none"> <li>• 25 tests/kit</li> <li>• <b>Vitassay Crypto+Giardia</b></li> <li>• Instructions for use.</li> <li>• 25 vials with diluent for the sample dilution.</li> </ul>	<ul style="list-style-type: none"> <li>• Specimen collection container.</li> <li>• Disposable gloves.</li> <li>• Timer.</li> </ul>

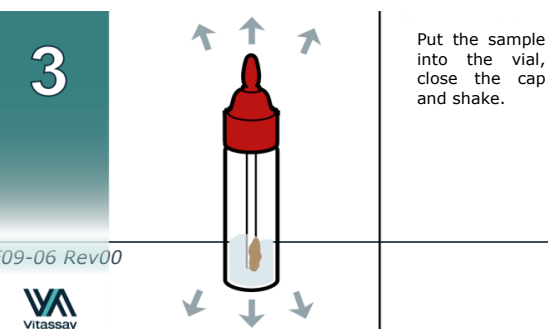
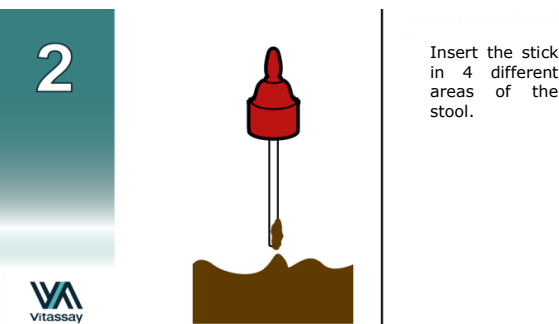
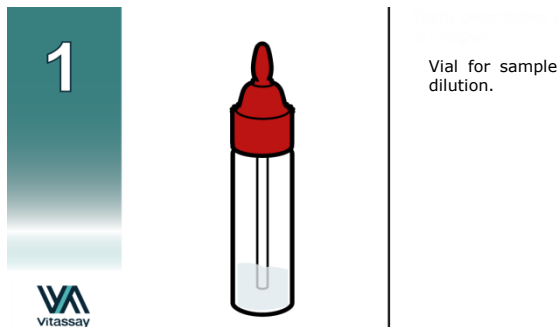
### SPECIMEN COLLECTION

Collect sufficient quantity of feces: 1-2g or mL for liquid samples. Stool samples should be collected in clean and dry containers.

Samples can be stored in the refrigerator (2-8°C/35.6-46.4°F) for 1-2 days prior to testing. For longer storage, maximum 1 year, the specimen must be kept frozen at -20°C (-4°F). Samples must be brought to room temperature before testing.

## SPECIMEN PREPARATION

1. Remove the cap of the vial with diluent for the sample dilution (figure 1).
2. Use the stick to collect sufficient sample quantity (approx. 125mg). For solid stool, insert the stick in 4 different areas of the stool sample taken approx. 125mg, (figure 2), and add it into the vial with diluent for the sample dilution. For liquid stool, take 125 µL of the sample using a micropipette and transfer it into the vial with diluent for the sample dilution.
3. Close the tube with the diluent and stool sample. Shake vigorously the vial in order to assure good sample dispersion (figure 3).

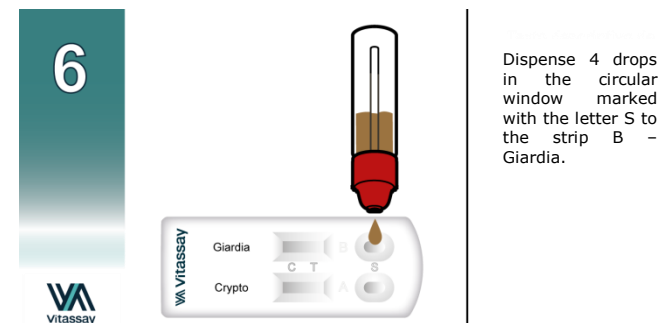
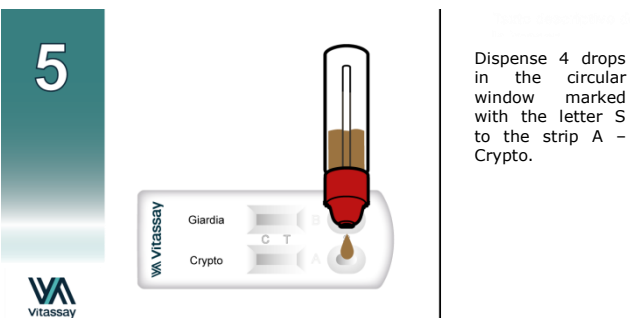
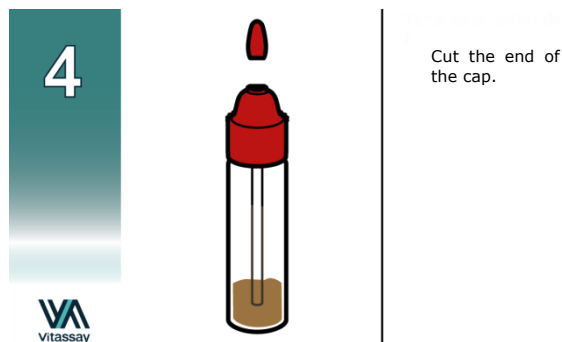


## PROCEDURE

Allow the test, stool sample, controls and diluent to reach room temperature (15-30°C/59-86°F) prior to testing. Do not open pouches until the performance of the assay.

1. Shake the vial with the sample vigorously to obtain a good sample dilution.
2. Remove the **Vitassay Crypto+Giardia** from its sealed bag just before using it.
3. Take the vial containing the diluted sample, cut the end of the cap (figure 4) and dispense 4 drops in the circular window marked with the letter A – Crypto (figure 5) and 4 drops, using the same vial, in the circular window marked with the letter B – Giardia (figure 6).
4. Read the results at **10 minutes**. Do not read the results later than 10 minutes.

If the test does not run due to solid particles, stir the sample added in the sample window with the stick. If it does not work, dispense a drop of diluent until seeing the liquid running through the reaction zone.



## INTERPRETATION OF THE RESULTS

RESULTS	Strip A Crypto	Strip B Giardia	INTERPRETATION
	NEGATIVE GREEN	NEGATIVE GREEN	There is no Cryptosporidium or Giardia presence. No infection caused by Cryptosporidium or Giardia.
	POSITIVE GREEN-RED	POSITIVE GREEN-RED	There is Cryptosporidium and Giardia presence. Infection caused by Cryptosporidium and Giardia.
	POSITIVE GREEN-RED	NEGATIVE GREEN	There is Cryptosporidium presence. Infection caused by Cryptosporidium.
	NEGATIVE GREEN	POSITIVE GREEN-RED	There is Giardia presence. Infection caused by Giardia.

<b>ANY OTHER RESULTS</b>	Invalid result, we recommend repeating the assay using the sample with another test. <b>Note:</b> Wrong procedural techniques or deterioration of the reagents are the main reasons of control line failure. If the symptoms or situation still persist, discontinue using the test kit and contact your local distributor.
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**Notes:** The intensity of the red colored test line in the result line zone (T) will vary depending on the concentration of antigens in the specimen.

**QUALITY CONTROL**

Internal procedural controls are included in **Crypto+Giardia**. Green lines appearing in the in the results window are internal controls, which confirm sufficient specimen volume and correct procedural technique.

**LIMITATIONS**

- **Vitassay Crypto+Giardia** must be carried out within 2 hours of opening the sealed bag.
- An excess of stool sample could cause wrong results (brown bands appear). Dilute the sample with the diluent and repeat the test.
- The intensity of test line may vary depending on the concentration of antigens.
- The use of other samples different from human fecal samples has not been established.
- After one week of infection, the number of parasites in feces is decreasing, making the sample less reactive. Stool samples should be collected within one week of the onset symptoms.
- The quality of **Vitassay Crypto+Giardia** depends on the quality of the sample; Proper fecal specimens must be obtained.
- Positive results determine the presence of Cryptosporidium and/or Giardia in faecal samples; nevertheless, a positive result should be followed up with additional laboratory techniques (biochemical methods or microscopy) to confirm the results. A confirmed infection should only be made by a physician after all clinical and laboratory findings have been evaluated and must be based in the correlation of the results with further clinical observations.
- A negative result is not meaningful because of it is possible the antigens concentration in the stool sample is lower than the detection limit values. If the symptoms or situation still persist, a

Cryptosporidium and/or Giardia determination should be carried out with another technique (for example microscopy).

**EXPECTED VALUES**

Infections are more often seen in young children as well as those who are immunocompromised. Enteric infections in children can have devastating consequences affecting intestinal absorption, nutrition, and childhood development. There is an increased risk of transmission on developing countries due to urban crowding and poor sanitation.

**PERFORMANCE CHARACTERISTICS**

**Clinical sensitivity and specificity**

An evaluation was performed using **Vitassay Crypto+Giardia** and these results were confirmed using a microscopy technique and PCR (positive results).

The Results were as follows:

IC test: Vitassay Crypto+Giardia	Microscopy technique/PCR		
	Positive	Negative	Total
	Positive	25	0
Negative	0	229	229
<b>Total</b>	25	229	254

Vitassay Crypto+Giardia (crypto) vs Microscopy technique			
Sensitivity	Specificity	PPV	NPV
>99%	>99%	>99%	>99%

IC test: Vitassay Crypto+Giardia	Microscopy technique/PCR		
	Positive	Negative	Total
	Positive	61	0
Negative	2	191	193
<b>Total</b>	63	191	254

Vitassay Crypto+Giardia (giardia) vs Microscopy technique			
Sensitivity	Specificity	PPV	NPV
97%	>99%	>99%	99%

The results showed that **Vitassay Crypto+Giardia** has a high sensitivity and specificity to detect Cryptosporidium and Giardia.

**Cross reactivity**







No cross reactivity was detected against other gastrointestinal pathogens that are occasionally present in feces:

<i>Campylobacter jejuni</i>	<i>Giardia lamblia (Strip A)</i>	<i>Salmonella typhimurium</i>
<i>Campylobacter coli</i>	<i>Helicobacter pylori</i>	<i>Shigella boydii</i>
<i>Clostridium difficile</i>	<i>Listeria monocytogenes</i>	<i>Shigella dysenteriae</i>
<i>Cryptosporidium parvum (Strip B)</i>	<i>Salmonella enteritidis</i>	<i>Shigella flexneri</i>
<i>Escherichia coli O157:H7</i>	<i>Salmonella paratyphi</i>	<i>Shigella sonnei</i>
<i>Entamoeba histolytica</i>	<i>Salmonella typhi</i>	<i>Staphylococcus aureus</i>

**REFERENCES**

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2. T. WEITZEL; S. DITTRICH; I. MÖHL; E. ADUSU; T. JELINEK. "Evaluation of seen commercial antigen detection test for Giardia and Cryptosporidium in stool samples". Clin Microbiol Infect 2006, 12: 656-659.
3. JULIANA MINAK; MAMUN KABIR; IQBAL MAHMUD; YUE LIU; LEI LIU; RASHIDUL HAQUE; WILLIAM A. PETRI JR. "Evaluation of Rapid Antigen Point-of-Care Test for Detection of Giardia and Cryptosporidium Species in Human Fecal Specimens". Journal of Clinical Microbiology, 2011, p. 154-156.

**SYMBOLS FOR IVD COMPONENTS AND REAGENTS**

<b>IVD</b>	in vitro diagnostic device		Keep dry
	Consult instructions for use		Temperature limitation
	Use by		Manufacturer
<b>LOT</b>	Batch code		Contains sufficient for <n> test
DIL	Sample diluent	<b>REF</b>	Catalogue number

