Datasheet for ABIN1305152

Giardia Lamblia ELISA Kit

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Overview

Quantity:	96 tests
Target:	Giardia Lamblia
Reactivity:	Giardia lamblia
Method Type:	Sandwich ELISA
Application:	ELISA

Product Details

Purpose:	This microplate-based ELISA (enzyme linked immunosorbent assay) kit is intended for the qualitative detection of Giardia lamblia antigen in feces. The assay is a useful tool in the diagnosis of active Giardia lamblia infection in acute or chronic gastroenteritis.
Sample Type:	Fecal
Analytical Method:	Qualitative
Detection Method:	Colorimetric
Specificity:	The assay does not cross react to following organisms: Cryptosoridium parvum, Rotavirus, Adenovirus.
Characteristics:	Gastrointestinal Disease
Components:	1. Anti-Giardia Antibody Coated Microplate. One bottle contains 30 mL of 20-fold concentrated buffer matrix with protein stabilizers and preservative. This reagent should be stored at 2-8 °C and is stable until the expiration date on the kit box. Before use the concentrated buffer must be diluted with 570 mL of distilled water and mixed well. Upon dilution this yields a working patient sample diluent containing a surfactant in phosphate-buffered saline with a non-azide

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	preservative. The diluted reagent can be stored at room temperature and is stable for 8 weeks. It can also be stored at 2-8 °C and is stable until the expiration date on the kit box.
Material not included:	1. Precision single channel pipettes capable of delivering 10 µL, 50 µL, 100 µL, and 1000 µL, etc. 2. Repeating dispenser suitable for delivering 100 µL. 3. Disposable pipette tips suitable for above volume dispensing. 4. Disposable 12 x 75 mm or 13 x 1 ? Repeating dispenser suitable for delivering 100 µL. ? Disposable pipette tips suitable for above volume dispensing. ? Disposable 12 x 75 mm or 13 x 100 glass or plastic tubes. ? Disposable plastic 1000 mL bottle with cap. ? Aluminum foil. ? Deionized or distilled water.? Plastic microtiter well cover or polyethylene film. ? ELISA multichannel wash bottle or automatic (semi-automatic) washing system. ? Spectrophotometric microplate reader capable of reading absorbance at 450 nm.

Target Details

Target:	Giardia Lamblia
Abstract:	Giardia Lamblia Products
Target Type:	Parasite
Background:	Giardia lamblia (also known as Giardia intestinalis) has a characteristic tear-drop shape and measures 10-15 µm in length. It has twin nuclei and an adhesive disk which is a rigid structure reinforced by supelicular microtubules. There are two median bodies of unknown function, but their shape is important for differentiating between species. There are 4 pairs of flagella, one anterior pair, two posterior pairs and a caudal pair. These organisms have no mitochondria, endoplasmic reticulum, golgi, or lysosomes. Giardia has a two-stage life cycle consisting of trophozoite and cyst. The life cycle begins with ingested cysts, which release trophozoites (10-20 µm x 5-15 µm) in the duodenum. These trophozoites attach to the surface of the intestinal epithelium using a ventral sucking disk and then reproduce by binary fission. The trigger for encystment is unclear, but the process results in the inactive, environmentally resistant form of Giardia -- a cyst (11-14 µm x 7-10 µm) that is excreted in feces. Giardiasis is a diarrheal illness caused by Giardia lamblia, after ingestion of Giardia cysts. Once a person has been infected with Giardia, the parasite lives in the intestine and is passed in the stool. Millions of germs can be released in a bowel movement from an infected human or animal. Giardia is found in soil, food, water, or surfaces that have been contaminated with the feces from infected humans or animals. Because the parasite is protected by an outer shell, it can survive outside the body and in the environment for long periods of time. Because it is spread world-wide, Giardia lamblia has

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	become one of the most important causes of chronic diarrheas. About 15-20 % of children under age ten years and 19 % of male homosexuals have been infected. Giardia infection can cause a variety of intestinal symptoms either acute or chronic, which include diarrhea, gas or flatulence, greasy stools that tend to float, stomach cramps, upset stomach or nausea. These symptoms may lead to weight loss and dehydration. Some people with giardiasis have no symptoms at all. Those asymptomatic cases still shed Giardia cycts. Generally, symptoms of giardiasis begin 1 to 2 weeks after becoming infected and they may last 2 to 6 weeks. The method used for the diagnosis of giardiasis in the past has been the detection of Giardia cysts in stool by microscopy. Recently, specific Giardia antigen ELISA greatly simplified the diagnostic procedure and is as sensitive as the microscopic method. Another advantage of using Giardia antigen ELISA is that it does not require the intact organisms in the test specimen.

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Sample Volume:	0.1 mL
Assay Time:	4 h
Plate:	Pre-coated
Protocol:	This sandwich ELISA is designed, developed and produced for the qualitative measurement of Giardia lamblia antigen in stool specimen. The assay utilizes the microplate-based enzyme immunoassay technique by coating highly purified antibody onto the wall of microtiter wells Assay controls and fecal specimen are added to microtiter wells of microplate that was coated with a highly purified polyclonal anti-Giardia lamblia antibody on its wall. The Giardia lamblia antigen will be bound to the antibody coated plate after an incubation period. The unbound matrices are washed away and a HRP-conjugated monoclonal antibody which specifically recognizes the protein of Giardia lamblia is added for further immunoreactions. After an incubation period, an immunocomplex of Anti-Giardia Antibody Giardia lamblia Antigen HRP-conjugated Anti-Giardia Tracer Antibody is formed if Giardia lamblia antigen is present in the test sample. The unbound tracer antibody and other protein or buffer matrix are removed in the subsequent washing step. HRP conjugated tracer antibody bound to the well is then incubated with a substrate solution in a timed reaction and then measured in a spectrophotometric microplate reader. The enzymatic activity of the tracer antibody bound to giardia proteins captured on the wall of each microtiter well is directly proportional to the amount of Giardia lamblia antigen level in each test specimen.
Reagent Preparation:	(1) Prior to use allow all reagents to come to room temperature. Reagents from different kit lot

	numbers should not be combined or interchanged. (2) Concentrated Patient Sample Diluent must be diluted to working solution prior use. Please see REAGENTS section for details.
Sample Collection:	Fresh fecal sample should be collected by using a plastic sampling device, for example, Epitope Diagnostics Fecal Sample Collection Device. It is required to collect a minimum of 0.1 mL liquid stool sample or 0.1 g solid sample. The collected fecal sample must be transported, kept at 2-8 °C and tested within 2 days. A non-preserved sample must be stored below -20 °C for a longer storage period. Fecal sample collected in 10 % formalin or SAF can be stored at 2-25 °C for 2 months.
Sample Preparation:	(1) Label a test tube (12x75 mm) or a 1.5 mLplastic vial. (2) Add 1 mL of assay buffer to each tube or vial. (3) Add 100 µL of liquid stool sample to the above tube. (4) With solid stool sample, take an equivalent amount (about 50 100 mg) with a spatula or a disposable inoculation loop. Suspend the solid stool sample with 1 mL patient sample diluent and mix well on a vortex mixer. (5) Centrifuge the diluted fecal sample at 3000 rpm (800- 1500 g) for 10 15 minutes. The supernatant can be directly used in the assay. As an alternative to centrifuging, let the diluted samples sit and sediment for 15 minutes and take the clear supernatant for testing. Note: If the test procedure is performed on an automated ELISA system, the supernatant must be particle-free by centrifuging the sample.
Assay Procedure:	(1) Place a sufficient number of anti-Giardia antibody coated microwell strips in a frame to run giardia controls (Cat. 30192-30193) and unknown samples in duplicate. (2) Test Configuration (3) Add 100 µL of controls and diluted patient stool samples into each designated microwell. (4) Cover the plate with a plate sealer and also with aluminum foil to avoid exposure to light. (5) Incubate plate at room temperature for 1 hour (6) Prepare working anti-Giardia tracer antibody working solution by 1:21 fold dilution of the Anti-Giardia Tracer Antibody with the Tracer Antibody Diluent. For each strip, it is required to mix 1 mL of Tracer Antibody Diluent with 50 µL of Tracer Antibody in a clean test tube. (7) Remove the plate sealer. Decant the contents of each well. Wash each well 5 times by dispensing 350 µL to 400 µL of deionized or distilled water into each well and then completely aspirating the contents. Alternatively, an automated microplate washer can be used. (Note: The plate must not be washed with any ELISA wash buffer!) (8) Add 100 µL of above diluted tracer antibody working solution to each of the wells. (9) Cover the plate with one plate sealer and also with aluminum foil to avoid exposure to light. (10) Incubate plate at room temperature for 45 minutes.

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	(11) Remove the plate sealer. Decant the contents of each well. Wash each well 5 times by dispensing 350 µL to 400 µL of deionized or distilled water into each well and then completely aspirating the contents. Alternatively, an automated microplate washer can be used. (Note: The plate must not be washed with any ELISA wash buffer!) (12) Add 100 µL of ELISA HRP Substrate into each of the wells. (13) Cover the plate with aluminum foil to avoid exposure to light (14) Incubate plate at room temperature for 20 minutes (15) Remove the aluminum foil. Add 100 µL of ELISA Stop Solution into each of the wells. Mix gently (16) Read the absorbance at 450 nm within 10 minutes in a microplate reader. As an alternative, one can interpret the test results visually by using the color code card included in the kit.
Assay Precision:	The reproducibility of this assay is validated by measuring four samples (two negative and two positive) both in a single assay of 12-replicate determinations and in 6 different assays run on different dates. The results showed a consistent test results interpretation for all the samples.
Restrictions:	For Research Use only

Handling

Precaution of Use:	The reagents must be used in a laboratory and are for professional use only. Source material for reagents containing bovine serum albumin was derived in the contiguous 48 United States. It was obtained only from healthy donor animals maintained under veterinary supervision and found free of contagious diseases. Wear gloves while performing this assay and handle these reagents as if they are potentially infectious. Avoid contact with reagents containing TMB, hydrogen peroxide, or sulfuric acid. TMB may cause irritation to skin and mucous membranes and cause an allergic skin reaction. TMB is a suspected carcinogen. Sulfuric acid may cause severe irritation on contact with skin. Do not get in eyes, on skin, or on clothing. Do not ingest or inhale fumes. On contact, flush with copious amounts of water for at least 15 minutes. Use Good Laboratory Practices.
Storage:	4 °C