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Datasheet for ABIN2014341  
**Helicobacter Pylori ELISA Kit**

1 Image

### Overview

Quantity:	96 tests
Target:	Helicobacter Pylori (H. pylori)
Reactivity:	Helicobacter pylori
Method Type:	Sandwich ELISA
Detection Range:	0.165-150 ng/mL
Minimum Detection Limit:	0.165 ng/mL
Application:	ELISA

### Product Details

Purpose:	This microplate-based ELISA (enzyme linked immunosorbent assay) kit is intended for the quantitative detection of Helicobacter pylori antigen in feces. The assay is a useful tool in the detection of active H. pylori infection.
Sample Type:	Fecal
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Cross-Reactivity (Details):	The assay does not cross react to the following organisms: Cryptosporidium parvum, Giardia lamblia, rotavirus and adenovirus.
Components:	1. H. pylori Antibody Coated Microplate. One microplate with 8 x 12 strips (96 wells total) coated with highly purified H. pylori antibody. The plate is framed and sealed in a foil zipper bag with a desiccant. This reagent should be stored at 2-8 °C and is stable until the expiration date on the kit box.

2. Anti-H.pylori Tracer Antibody. One vial containing 12 mL ready-to-use horseradish peroxidase (HRP) -conjugated monoclonal H. pylori antibody in a stabilized protein matrix. This reagent should be stored at 2-8 °C and is stable until the expiration date on the kit box.
3. ELISA HRP Substrate. One bottle containing 12 mL of tetramethylbenzidine (TMB) with hydrogen peroxide. This reagent should be stored at 2-8 °C and is stable until the expiration date on the kit box.
4. ELISA Stop Solution. One bottle containing 12 mL of 0.5 M sulfuric acid. This reagent should be stored at 2-8 °C or room temperature and is stable until the expiration date on the kit box.
5. H. pylori Antigen Calibrator Level 6. 1 vial containing 1.5 mL of H.pylori Antigen Calibrator Level 6. This calibrator is in a liquid bovine serum albumin-based matrix with mercury and sodium azide preservative. Refer to vials for exact concentration. This reagent should be stored at 2-8 °C and are stable until the expiration date on the kit box, -2 °C for long term storage.
6. ELISA Wash Concentrate. One bottle containing 3 mL of 30-fold concentrate. Before use the contents must be diluted with 87 mL of distilled water and mixed well. Upon dilution this yields a working wash solution containing a surfactant in phosphate buffered saline with a non-azide and non-mercury based preservative. The diluted wash buffer should be stored at room temperature and is stable until the expiration date on the kit box.
7. H. Pylori Concentrated Assay Buffer. One bottle containing 3 mL of 4-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 90 mL of demineralized water and mixed well. Upon dilution, this yields as 1x Assay Buffer, which serves as a Calibrator Level 1, and as a patient sample diluent containing a surfactant in phosphate-buffered saline with a non-azide preservative. The diluted reagent is stored at 2-8 °C and is stable until the expiration date on the kit box

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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 100 glass or plastic tubes.
5. Disposable plastic 1000 mL bottle with cap.
6. Aluminum foil.
7. Deionized or distilled water.
8. Plastic microtiter well cover or polyethylene film.
9. ELISA multichannel wash bottle or automatic (semi-automatic) washing system.
10. Spectrophotometric microplate reader capable of reading absorbance at 450/2. nm.

## Target Details

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Target:	Helicobacter Pylori (H. pylori)
Alternative Name:	Helicobacter Pylori ( <a href="#">H. pylori Products</a> )
Target Type:	Virus
Background:	H. pylori (previously known as Campylobacter pyloridis) is a type of bacteria that infects the stomach and is a common cause of peptic ulcers. H. pylori bacteria can be passed from person to person through direct contact with saliva, vomit or fecal matter. H. pylori can also be spread through contaminated food or water. The infection is normally acquired during childhood. H. pylori usually goes undiagnosed until symptoms of a peptic ulcer occur. H. pylori infection is quite common and is present in about half the people in the world.

## Application Details

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Sample Volume:	1.5 mL
Assay Time:	4 h
Plate:	Pre-coated
Protocol:	<p>This sandwich ELISA is designed, developed and produced for the quantitative measurement of H. pylori 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 calibrators and fecal specimen are added to microtiter wells of microplate that was coated with a highly purified monoclonal H. pylori antibody on its wall. During the assay, the H. pylori antigen will be bound to the antibody coated plate after an incubation period. The unbound material is washed away and another HRP-conjugated monoclonal antibody which specifically recognizes the protein of H. pylori is added for further immunoreactions. After an incubation period, the immunocomplex of H. pylori Antibody H. pylori Antigen HRP-conjugated Anti-H. pylori Tracer Antibody is formed if H. pylori antigen is present in the test sample. The unbound tracer antibody and other proteins in 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 H. pylori proteins captured on the wall of each microtiter well is directly proportional to the amount of H. pylori antigen level in each test specimen.</p>
Reagent Preparation:	<p>(1) Prior to use allow all reagents to come to room temperature. Reagents from different kit lot numbers should not be combined or interchanged.</p> <p>(2) Concentrated Assay Buffer must be diluted to working solution prior use. Please see</p>

REAGENTS section for details.

(3) ELISA Wash Concentrate must be diluted to working solution prior use. Please see REAGENTS section for details.

(4) Prepare 1:3 serially diluted calibrators using H.pylori Ag Calibrator Level 6 and 1x Assay Buffer as the dilution buffer. Store at 2-8 °C, -20 °C for long term storage. Avoid more than 3x freeze thaw cycle.

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**Sample Collection:** Fresh fecal sample should be collected into a stool sample collection container. It is required to collect a minimum of 1-2 mL liquid stool sample or 1-2g solid sample. The collected fecal sample must be transported to the lab in a frozen condition (-20 °C). If the stool sample is collected and tested the same day, it is allowed to be stored at 2-8 °C.

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**Sample Preparation:**

- (1) Label a test tube (12x75 mm) or a 4 mL plastic vial.
- (2) With solid stool sample, take or weigh an equivalent amount (about 40 mg, size as a grain of rice) with a spatula or a disposable inoculation loop. Suspend the solid stool sample with 1 mL 1x Assay Buffer and mix well on a vortex mixer.
- (3) Centrifuge the diluted fecal sample at 3000 rpm (800- 1500 g) for 5-10 minutes. The supernatant can be directly used in the assay. As an alternative to centrifuging, let the diluted samples sit and sediment for 30 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.
- (4) This sample can be stored at 2-8 °C up to three days and below -20 °C for longer storage. Avoid more than 3x freeze and thaw cycle.

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**Assay Procedure:**

- (1) Place a sufficient number of H. Pylori monoclonal antibody-coated microwell strips in a frame.
- (2) Test Configuration
- (3) Add 100 µL of calibrators and diluted patient stool samples into the designated microwell. Mix by gently tapping the plate. Cover the plate with one plate sealer. Cover with foil or other material to protect from light.
- (4) Incubate plate at room temperature for 1 hour.
- (5) Remove the plate sealer. Aspirate the contents of each well. Wash each well 5 times by dispensing 350 µL to 400 µL of working wash solution into each well, then completely aspirating the contents. Alternatively, an automated microplate washer can be used.
- (6) Add 100 µL ready-to-use anti- H. Pylori Tracer Antibody . Mix by gently tapping the plate.
- (7) Cover the plate with one plate sealer and also with aluminum foil to avoid exposure to light
- (8) Incubate plate at room temperature for 30 minutes.
- (9) Remove the plate sealer. Aspirate the contents of each well. Wash each well 5 times by

## Application Details

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- dispensing 350  $\mu$ L to 400  $\mu$ L of working wash solution into each well and then completely aspirating the contents. Alternatively, an automated microplate washer can be used.
- (10) Add 100  $\mu$ L of ELISA HRP Substrate into each of the wells.
- (11) Cover the plate with a new plate sealer and also with aluminum foil to avoid exposure to light.
- (12) Incubate plate at room temperature for 20 minutes.
- (13) Remove the aluminum foil and plate sealer. Add 100  $\mu$ L of ELISA Stop Solution into each of the wells. Mix gently.
- (14) Read the absorbance at 450 nm with reference filter at 620 nm or 650 nm.

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Calculation of Results:	<ol style="list-style-type: none"><li>1. Calculate the average absorbance for each pair of duplicate test results.</li><li>2. Subtract the average absorbance of the calibrator 1 (0 ng/mL) from the average absorbance of all other readings to obtain corrected absorbance.</li><li>3. The calibrator curve is generated by the corrected absorbance of all calibrator levels on the ordinate against the calibrator concentration on the abscissa using point-to-point or log-log paper. Appropriate computer assisted data reduction programs may also be used for the calculation of results. The H. Pylori concentrations for the unknown samples are read directly from the calibrator curve using their respective corrected absorbance</li></ol>
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Assay Precision:	The intra-assay precision is validated by measuring two samples in a single assay with 12 replicate determinations. The inter-assay precision is validated by measuring two samples in duplicate in 12 individual assays.
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Restrictions:	For Research Use only
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## Handling

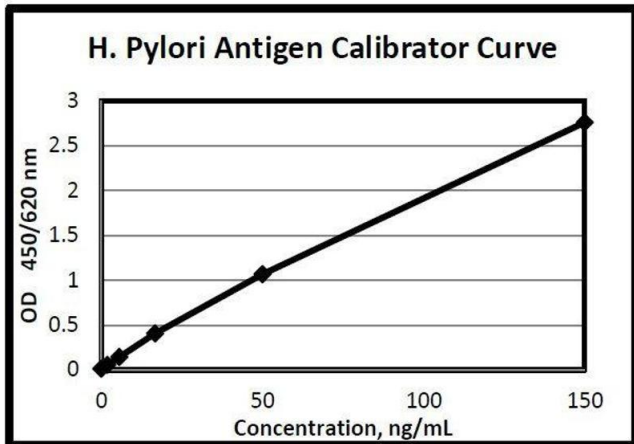
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Precaution of Use:	The reagents must be used in a laboratory and are for professional use only. Materials sourced for reagents containing bovine serum albumin were derived in the contiguous 48 United States and obtained only from healthy donor animals 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.
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Storage:	4 °C
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## ELISA

Image 1.