Willi Fox

H. Pylori Antigen in faeces Test

H. Pylori Antigen in faeces Test Package Insert

A rapid, one step test for the qualitative detection of H. Pylori Antigen in faeces.

For professional in vitro diagnostic use only.

1. Intended use

The Willi Fox - *H. pylori* Antigen Rapid Test Device (Faeces) is a rapid visual immunoassay for the qualitative presumptive detection of *Helicobacter pylori* antigens in human fecal specimens. This kit is intended to be used as an aid in the diagnosis of *H. pylori* infection.

2. Introduction

Helicobacter pylori (also known as Campylobacter pylori) is a spiral-shaped with a typical flagellum, Gram negative bacteria, infecting gastric mucosa. It causes several gastro-enteric diseases such as non-ulcerous dyspepsia, gastric and duodenal ulcer, active gastritis and can even increase the risk of stomach adenocarcinoma, so as to be classified as carcinogen agent type I.

Many *H. pylori* strains have been isolated: among them, the strain expressing CagA antigen is strongly immunogenic and, according to this, it is of utmost clinical importance because it is associated to the cytotoxic factor. It is widely reported in many literature *articles* that, in infected patients showing antibodies against CagA gene product, the risk of gastric cancer is up to five times higher than the reference group infected with a CagA negative bacterial strain.

The presence of the gene itself determines the persistence of the infection, the ulceration and the protein associated, VacA toxin is frequently the main cause of infiltrations in the gastric mucosa. This antigen associated to others, such as CagII, CagC, seems to act as starting agent of a sudden inflammatory response which can provoke ulceration (peptic ulcer), allergic episodes, and a decrease of the therapy efficacy.

At present several invasive and non-invasive approaches are available to detect this infection state. Invasive methodologies requires endoscopy of the gastric mucosa with a histologic, cultural and urease investigation, which are cost-effective and requiring long times to come to a correct final diagnosis. Alternatively, non-invasive methods are available such as Breath Test, which is extremely complicated and not highly selective, or classical ELISA and immunoblotting assays.

3. Principle

The *Willi Fox* - *H. pylori* Antigen Rapid Test Device (Faeces) has been designed to detect *Helicobacter pylori* through visual interpretation of color development in the internal strip. The membrane was immobilized with anti-*H. pylori* monoclonal antibody on the test region. During the test, the specimen is allowed to react with colored anti-*H. pylori* monoclonal antibody colloidal gold conjugates, which were precoated on the sample pad of the test. The mixture then moves on the membrane by a capillary action, and interact with reagents on the membrane. If there were enough *H. pylori* antigens in specimens, a colored band will form at the test region of the membrane. Presence of this colored band indicates a positive result, while its absence indicates a negative result. Appearance of a colored band at the control region serves as a procedural control. This indicates that proper volume of specimen has been added and membrane wicking has occurred.

4. Kit contents

Test Kit Contents:

- *H. pylori* Antigen Test Devices
- Sample collection tube with buffer
- Desiccant

5. Material required but not provided

- Timer
- Specimens collection container
- Centrifuge

7. Precautions

- For professional *in vitro* diagnostic use only.
- Do not use after the expiration date indicated on the package. Do not use the test if the foil pouch is damaged. Do not reuse tests.
- This kit contains products of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not completely guarantee the absence of transmissible pathogenic agents. It is therefore, recommended that these products be treated as potentially infectious, and handled by observing usual safety precautions (e.g., do not ingest or inhale).
- Avoid cross-contamination of specimens by using a new specimen collection container for each specimen obtained.
- Read the entire procedure carefully prior to testing.
- Do not eat, drink or smoke in the area where the specimens and kits are handled. Handle all specimens as if they contain infectious agents. Observe established precautions against microbiological hazards throughout the procedure and follow standard procedures for the proper disposal of specimens. Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are assayed.
- Humidity and temperature can adversely affect results.
- Used testing materials should be discarded according to local regulations.

8. Storage and stability

The kit should be stored at 2-30℃ until the expiry date printed on the sealed pouch.

- The test must remain in the sealed pouch until use.
- **Do not freeze.**
- Care should be taken to protect the components of the kit from contamination. Do not use if there is evidence of microbial contamination or precipitation. Biological contamination of dispensing equipments, containers or reagents can lead to false results.

9. Specimen collection and preparation

- The *H. pylori* Antigen Rapid Test Device (Feces) is intended only for use with human fecal specimens.
- Perform the testing immediately after the specimen collection. Do not leave the specimens at room temperature for prolonged periods. Specimens may be stored at 2-8℃ for up to 72 hours.
- Bring specimens to room temperature prior to testing.
- Pack the specimens in compliance with applicable regulations for transportation of etiological agents, in case they need to be shipped.

10. Procedure

Bring tests, specimens, buffer and/or controls to room temperature (15-30℃) before use.

1. Specimen collection and pre-treatment:

1) Best results will be obtained if the assay is performed within 6 hours after collection.

2) Unscrew and remove the dilution tube applicator. Be careful not to spill or spatter solution from the tube. Collect specimens by inserting the applicator stick into at least 3 different sites of the feces to collect approximately 50 mg of feces (equivalent to 1/4 of a pea).

3) Place the applicator back into the tube and screw the cap tightly. Be careful not to break the tip of the dilution tube.

4) Shake the specimen collection tube vigorously to mix the specimen and the extraction buffer.

2. Testing

1) Remove the test from its sealed pouch, and place it on a clean, level surface. Label the test with patient or control identification. To obtain a best result, the assay should be performed within one hour.
2) Using a piece of tissue paper, break the tip of the dilution tube. Hold the tube vertically and dispense 2 drops of solution into the specimen well (S) of the test device.

Avoid trapping air bubbles in the specimen well (S), and do not drop any solution in observation window.

As the test begins to work, you will see color move across the membrane.

3. Wait for the colored band(s) to appear. The result should be read at 10 minutes. Do not interpret the result after 20 minutes.

Note: If the specimen does not migrate (presence of particles), centrifuge the extracted specimens contained in the extraction buffer vial. Collect 80 µL of supernatant, dispense into the specimen well (S) of a new test device and start afresh following the instructions mentioned above

11. Interpretation of results

POSITIVE*: Two distinct colored lines appear.

One line should be in the control line region (C) and another line should be in the test line region (T).

***NOTE:**

The intensity of color in the test region (T) may vary depending on the concentration of analytes present in the specimen. Therefore, any shade of color in the test region should be considered positive. Note that this is a qualitative test only, and cannot determine the concentration of analytes in the specimen.

NEGATIVE: One colored line appears in the control line region(C).

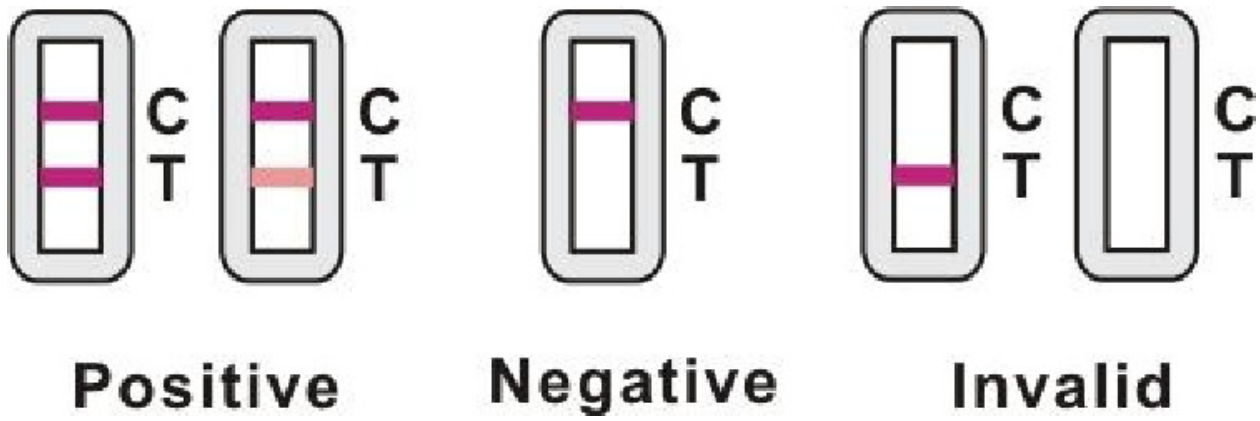
No apparent colored line appears in the test line region (T).

INVALID*: Control line (C) fails to appear.

Results from any test which has not produced a control band at the specified read time must be discarded. Please review the procedure and repeat with a new test. If the problem persists, discontinue using the kit immediately and contact your local distributor.

***NOTE:**

Insufficient specimen volume, incorrect operating procedure or expired tests are the most likely reasons for control band failure.



12. Quality control

- Internal procedural controls are included in the test. A colored band appearing in the control region is considered an internal positive procedural control, confirming sufficient specimen volume and correct procedural technique.
- External controls are not supplied with this kit. 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.

13. Limitations

1. The *Willi Fox* - *H. pylori* Antigen Rapid Test Device (Feces) is for professional *in vitro* diagnostic use, and should be used for the qualitative detection of *Helicobacter pylori* only.
2. Following certain antibiotic treatments, the concentration of *H. pylori* antigens may decrease to the concentration below the minimum detection level of the test. Therefore, diagnosis should be made with caution during antibiotic treatment.
3. As with all diagnostic tests, a definitive clinical diagnosis should not be based on the results of a single test, but should only be made by the physician after all clinical and laboratory findings have been evaluated.

14. Performance characterstics

Table: *Willi Fox* - *H. pylori* Antigen Rapid Test vs. Endoscope-based methods

Relative Sensitivity:
>99.9% (97.3%-100.0%)*

Relative Specificity:
>99.9% (97.6%-100.0%)*

Overall Agreement:
>99.9% (98.7%-98.8%)*

***95% Confidence Interval**

		***Willi Fox* - *H. pylori* Antigen Rapid Test**		
		+	**-**	**Total**
Biopsy/ Hystology/ RUT	**+**	132	0	132
	-	0	154	154
		132	154	286

Specificity:

Cross reactivity with following organisms has been studied at 1.0 x 10^9 organisms/ml. The following organisms were found negative when tested with the *Willi Fox* - One Step H. pylori Antigen Test Device (Feces):

Staphylococcus aureus	*Proteus mirabilis*	*Neisseria gonorrhea*
Pseudomonas aeruginosa	*Acinetobacter spp*	*Group B Streptococcus*
Enterococcus faecalis	*Salmonella choleraesius*	*Proteus vulgaris*
Group C Streptococcus	*Gardnerella vaginalis*	*Enterococcus faecium*
Klebsiella pneumoniae	*Acinetobacter calcoaceticus*	*Hemophilus influenzae*
Branhamella catarrhalis	*E.coli*	*Neisseria meningitidis*
Candida albicans	*Chlamydia trachomatis*	*Rotavirus*

15. Bibliography

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4. Cutler AF. Testing for Helicobacter pylori in clinical practice. Am j. Med. 1996; 100:35S-41S.
5. Anand BS, Raed AK, Malaty HM, et al. Loe point prevalence of peptic ulcer in normal individual with Helicobacter pylori infection. Am J Gastroenterol. 1996,91:1112-1115.

16. Glossary of symbols

REF	Catalog number		Do not reuse
LOT	Batch code		Use by
	Temperature limitation		Content
IVD	*In vitro* diagnostic medical device		Consult instructions for use
	Manufacturer		

CE

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