P-5242-B

BioSign® *H. pylori* WB

New One-Step Anti-*H. pylori* Antibody Test

For Professional *In Vitro* Use

Immunoassay for the Qualitative Detection of Anti-*Helicobacter pylori* Antibody in Human Whole Blood, Serum or Plasma

CLIA Complexity: Moderate
CDC Analytic ID Code: 2513

Catalog No.	BSP-406WB-35	35 Test Kit
	BSP-406WB-10	10 Test Kit

Intended Use

BioSign® *H. pylori* WB qualitatively detects total anti-*H. pylori* antibody in human whole blood, serum or plasma specimens. The test is intended for use in the diagnosis of *Helicobacter pylori* infection in adult patients with symptoms of gastrointestinal disorders.

Summary and Explanation

Helicobacter pylori, formerly known as *Campylobacter pylori*, are gram-negative microaerophilic spiral bacteria that have been identified and cultured since the past decade.[1] They are able to colonize the gastric mucosa for years[2], and their presence is strongly associated with chronic, diffuse and superficial gastritis of the fundus and antrum.[3-5] As a result, they are now believed to have an etiologic role in gastritis.[6,7] Recent evidence suggests that H. pylori gastritis may progress over several decades to chronic atrophic (type B) gastritis[8,9], a lesion that is a precursor of gastric carcinoma. The epidemiological features of gastric carcinoma and *H. pylori* infection are similar[10], and recent studies suggest that *H. pylori* infection may be a risk factor for gastric carcinoma.[11,12]

Until recently, diagnosis of infection with *H. pylori* required endoscopy and identification of the organism by means of subsequent culture of the bacteria and/or recognition of spiral organisms in histologically evaluated sections of gastric tissue. However, the expense and invasive nature of this procedure make endoscopy impractical for epidemiologic studies. Therefore, serologic test has been commonly used for the epidemiologic studies. There is an excellent correlation between a classical clinical presentation of gastritis, the presence of *H. pylori* in the stomach and elevated serum levels of anti-*H. pylori* antibodies.[13-15] Positive results can justify a short empirical trial of antimicrobial therapy in gastritis of unknown origin. Furthermore response to treatment can be serially monitored because levels of *H. pylori*-specific antibodies can be expected to fall significantly after successful antibacterial therapy.[16]

Principle

The **BioSign® *H. pylori* WB**—One-Step Anti-*H. pylori* Antibody Test utilizes solid-phase immunoassay technology for the qualitative detection of *H. pylori* antibodies in human whole blood, serum or plasma. In the test procedure, 25 µL of whole blood or 10 µL of serum or plasma sample is spotted in the Sample well (S), located below the Result window. The Developer solution is then added in the Sample well. The solution mobilizes the dye conjugated to *H. pylori* antigen and to anti-human immunogloburin antibodies. If any anti-*H. pylori* antibody is present in the sample, the dye conjugate will bind to the *H. pylori* antigen band impregnated in the test membrane. Visualization of the antigen band in the Test position (T) will occur only when the anti-*H. pylori* antibody is present in the sample. As the antibody-dye conjugate continues to move along the test membrane, it will be captured by a species specific antibody located in the Control position (C) to generate a colored band regardless of the presence of *H. pylori* antibodies in the sample. Therefore, the presence of two colored bands, one in the Test position and the other in the Control position, indicates a positive result, while the absence of a colored band in the Test position indicates a negative result.

Reagents and Materials Provided

- Test devices in sealed pouch.
- Capillary tubes (25 µL) for whole blood specimen (optional)
- A dropper bottle of Developer solution.
- Directions for Use.

Materials Required But Not Provided

- Vacutainer tubes for either serum or plasma procedure
- Centrifuge
- Micropipette (0–200 µL range)

Warnings and Precautions

- For *in vitro* diagnostic use only.
- Do not use kit beyond the expiration date.
- Observe proper precautions during handling specimens and follow the standard procedures for proper disposal of specimens.
- Upon disposal of the Developer solution, flush with a large volume of water to prevent azide buildup in drains.
- Do not use the test if the pouch is damaged.

Storage and Stability

Store kit at 2–30°C (35–86°F) in the original sealed pouch. The kits are stable until the expiration date.

Specimen Collection and Preparation

- **Anticoagulated Whole Blood**: Whole blood collected over heparin, citrate or EDTA can be used. Mix whole blood by inversion and use in the test as outlined in the Test Procedure. If testing cannot be performed within 24 hours, separate plasma and store at or below –20°C.
- **Fingertip Whole Blood**: Prick the finger and collect the blood in a capillary tube to the 25 µL mark. Transfer the blood onto the Sample well (**S**) of the test device and follow the Test Procedure.
- **Serum**: Allow the blood to clot, then centrifuge to separate the serum.

- **Plasma**: Collect the whole blood sample into a tube containing anticoagulant such as heparin, citrate or EDTA. Centrifuge the blood and separate the plasma.

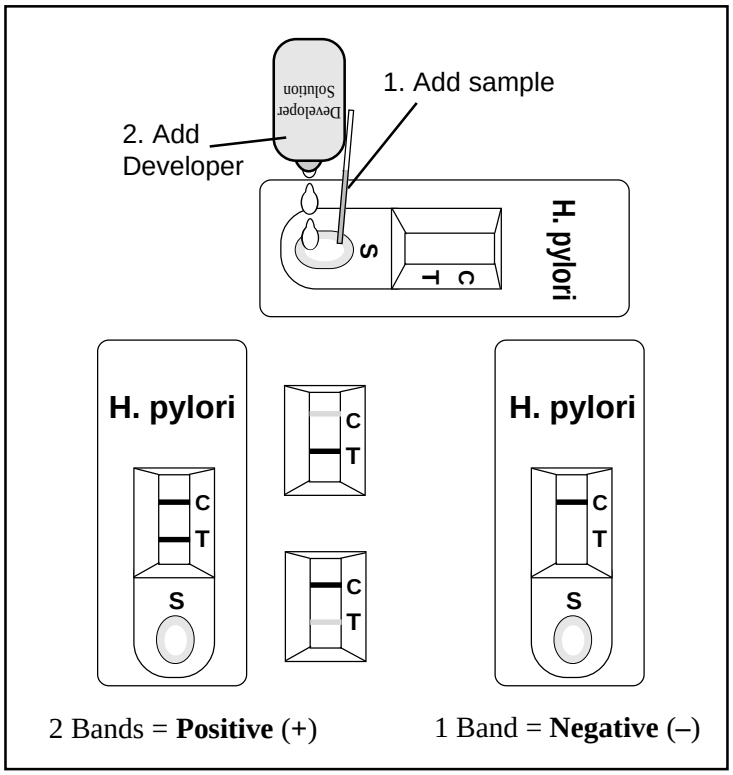
Procedure

Procedural Notes

- Allow specimens and the test kit to warm to room temperature (18–30°C) before testing.
- Do not open the sealed pouch until ready to perform the test.
- Several tests may be run at a time.
- Use a new capillary tube or micropipette tip for each specimen.
- Label the device with the patient's name or control number.
- Dispense the specimen at the upper area of the Sample well (S).
- Hold the dropper bottle in a vertical position and add 2 to 3 drops onto the lower area of the Sample well.
- Do not read the test result 15 minutes after the addition of Developer solution.
- After testing, dispose of the device and the used tip following good laboratory practices.

Test Procedure

1. Add 25 µL of whole blood using a capillary tube, or 10 µL of serum or plasma using a micropipette onto the upper area of the Sample well (see figure below).
2. Add 2 to 3 drops of Developer solution onto the lower area of Sample well (**S**).
3. Read test results at 10 minutes after the addition of Developer solution.



Interpretation of Results

Positive

One colored band each in the Test position (**T**) and in the Control position (**C**) means that antibodies against *H. pylori* have been detected.

Note: The test result can be read as soon as a distinct pink-purple colored line in the Test position (**T**) and a colored line in the Control position (**C**) appear. Any light to dark pink-purple colored line in the Test position should be read as a positive. The three possible positive cases are:

a. Two strong colored bands in both the Test (**T**) and Control (**C**) positions.

b. One strong colored band in the Test position (**T**) and one light colored band in the Control position(**C**).

c. One light colored band in the Test position (**T**) and one strong colored band in the Control position (**C**).

Negative

One colored band in the Control position (**C**) with no colored band in the Test position (**T**) means that antibodies against *H. pylori* have not been detected.

Invalid

The test is invalid if no band forms in the Control position. Repeat the test with a new test device.

Limitations

- The results obtained by this kit should be used only to evaluate patients with other clinical symptoms of gastrointestinal disease. This assay is not intended for use with asymptomatic patients.
- The performance characteristics of this test with specimens from pediatric patients have not been established.
- A positive result only means the presence of antibodies to *H. pylori* and does not indicate any disease status of patient. A positive test result does not allow one to distinguish between active infection and colonization by *H. pylori*.
- A negative result suggests that antibodies to *H. pylori* is not present, or is present at a level below the detection limit. If the test result is negative and infection of *H. pylori* is suspected, additional testing such as culture and histological analysis is recommended.

User Quality Control

- A quality control check is recommended using commercially available control sera. The frequency of Q.C. tests is determined according to your laboratory's standard Q.C. procedures. Upon confirmation of the expected results, the kit is ready for use with patient specimens. If external controls do not perform as expected, do not use the test kits. Repeat the test or contact PBM Technical Assistance.
- When the test has been performed correctly and the device is working properly, a distinct colored line will always appear in the Control position (**C**). The colored line in the Control position (**C**) is considered an internal positive procedural control. If the line does not appear, a new device should be tested. If the problem persists, contact PBM Technical Services.
- When the test has been performed correctly and the device is working properly, the background in the Result window will clear, providing a distinct test result. This clearing background in the Result window is considered an internal negative procedural control.

Expected Values

1. *H. pylori* is detectable in nearly 100% of adult patients with duodenal ulcer and about 80% of patients with gastric ulcer.[13,17] **BioSign® *H. pylori* WB** demonstrated positive results on 94% of patients with a symptom of ulcer and positive results on 80% of gastritis patients.
2. The prevalence of *H. pylori* antibody increases with age, is on the order of 5% in children, about 33% in randomly chosen blood donors, and approaches 50% at age 60 in the normal population of industrialized nations.[16,18] More than 25% of these infected patients are totally asymptomatic. Other factors such as socioeconomic status, ethnic group, different populations, geographical location and the type of the trouble associated with the infection also contribute to the observed variations in prevalence.
3. Asymptomatic and untreated patients continue to test IgG seropositive as long as the *H. pylori* organisms are present, even after histological resolution.[16] Hence, positive results are simply consistent with the diagnosis of *H. pylori*-associated gastritis or duodenal ulcer; whereas, negative results are strong evidence against these diagnoses

Performance Characteristics

Clinical specimens were collected from 207 symptomatic and asymtomatic individuals who presented for endoscopic examination. The age range was 19-83 years with a mean age of 52 years. The performance characteristics of **BioSign® *H. pylori* WB** were evaluated by comparison to biopsy/histology, agglutination test and ELISA for detection of anti-*H. pylori* antibody. The results are summarized in tables below.

Table 1. **BioSign® *H. pylori* WB** Test Result versus Biopsy/Histology

		Biopsy/Histology		
		Positive	Negative	Total
BioSign® *H. pylori* WB	Positive	71	14	85
	Negative	3	115	118
	Total	74	129	203

When biopsy/histology was used as a reference, the **BioSign® *H. pylori* WB** test demonstrated 95.9% sensitivity, 89.1% specificity and 91.6% agreement. Four tests were excluded in the calculation due to indeterminate result.

Table 2. **BioSign® *H. pylori* WB** Test Result versus Agglutination Test

		Agglutination test		
		Positive	Negative	Total
BioSign® *H. pylori* WB	Positive	80	8	88
	Negative	6	113	119
	Total	86	121	207

When the agglutination test was used as a reference, the **BioSign® *H. pylori* WB** test demonstrated 93.2% agreement.

Table 3. **BioSign® *H. pylori* WB** Test Result versus ELISA

		ELISA		
		Positive	Negative	Total
BioSign® *H. pylori* WB	Positive	78	10	88
	Negative	6	113	119
	Total	84	123	207

When the ELISA was used as a reference, the **BioSign® *H. pylori* WB** demonstrated 92.3% agreement.

Matrices Effect Study

Effect of specimen matrices on the result of **BioSign® *H. pylori* WB** test was evaluated using 59 matched specimen sets of each consisting of venous whole blood, capillary whole blood, plasma and serum in the **BioSign® *H. pylori* WB** test. Of 59 samples, 45 samples were positive and 13 samples were negative. Excellent agreement (>99%) was found between venous whole blood, capillary whole blood, plasma and serum indicating no significant effect of matrices on the test.

Reproducibility

Reproducibility of **BioSign® *H. pylori* WB** was evaluated by testing the negative, low positive and high positive samples. The samples were tested in replicates of 10 in a blind study by 4 technicians, on 3 different dates and at 4 different locations. The results showed 100% agreement with the expected results.

Proficiency Study

BioSign® *H. pylori* WB was evaluated at 3 different physicians' office laboratories using a panel of 90 coded samples. The proficiency panel contained negative, low positive and high positive specimens in either serum or whole blood. Either technical or non-technical personnel at three different institutions and three different days conducted the tests. The results obtained from 270 tests had a >99% agreement with the expected results. No significant differences were observed between the laboratories or personnel results.

Interference Study

Possible interference materials found in blood, such as, bilirubin, hemoglobin, triglycerides, or albumin, were tested in the **BioSign® *H. pylori* WB** test at approximately 10-fold higher concentrations. These substance did not alter the test results of **BioSign® *H. pylori* WB**.

References

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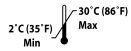
Symbols Key

	Manufactured by
CE	CE Mark
EC REP	Authorized Representative
IVD	*In Vitro* Diagnostic Medical Device
REF	Catalog Number
	Consult Instructions for Use
LOT	Batch Code
	"Use By" date in year-month-day format
	Temperature Limitation
	Contains sufficient for <n> tests
	Do not reuse
CONT	Contents
DEV	Test Device
SOL	Developer Solution
IFU	Instructions for Use
TEST HPY	Helicobacter Pylori Test

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