DyonMed S.A.
Point of Care Solutions

182-184 Lavriou Av., 15354, Peania, Attika
Tel. (+30) 210.66.55.494 & -495, Fax: (+30) 211.770.8599
info@dyonmedical.com www.dyonmedical.com
ISO 9001:2008 (No. 010110003)| ISO 13485:2003 (No 01210005)
by TÜV Austria Hellas

DyonHpAb®triple (*H. pylori* Ab IgG, IgM, IgA)

No. of CAT.: DNWA101

Rapid test for qualitative detection of antibodies IgG, IgM, IgA of Helicobacter pylori in samples of whole blood, capillary blood, serum or plasma

SUMMARY

DyonHpAb®triple (*H. pylori* Ab IgG, IgM, IgA) is a rapid immunochromatographic assay for the qualitative detection of human antibodies IgG, IgM, IgA against of Helicobacter pylori in samples of whole blood, capillary blood, serum or plasma.

Helicobacter pylori (known as *Campylobacter pylori*) is a spiral shaped, Gram-negative bacterium with flagella characteristics that affect the gastric mucosa, which may be parasite for years without necessarily causing symptoms. Unclear factors like stress, low pH or eating foods rich in spices may trigger the microbe resulting in multiple gastrointestinal diseases: gastric and duodenal ulcer, active gastritis, non-ulcerative dyspepsia, resulting in some cases in adenocarcinoma of stomach. The International Agency for Research on Cancer (IARC) and the World Health Organization (WHO) classify Helicobacter pylori (H. pylori) as a type 1 carcinogen and strictly recommended - constitute the identification and its treatment (Atherton and Blaser, 2009; Dorer et al, 2009).

Helicobacter pylori parasites around the planet and prevalence associated with age, genetic, economic, social and other factors (Atherton and Blaser, 2009). From 80% to 100% of patients with gastric ulcer are positive for the bacterium. Beyond the obvious emergency detection of these diseases, is directly recommended starting treatment (test and treat strategy) for confirmation of infection with rapid testing in special populations (Chey & Wong, 2007). Active treatment of the microbe with antibiotics seems to have led to the reduction of the prevalence of the virus in the U.S., and thus the simultaneous reduction in the frequency of hospitalization and hospital stay (Feinstein et al, 2010).

Detection of infection with Helicobacter pylori achieved with invasive and non-invasive approaches. Using methods of point of care, which detect antibodies against of Helicobacter pylori directly in blood samples, are widely used in clinical practice. These methods are advantageous point of care due to speed, convenience, accuracy and repeatability to use, unlike other non-invasive methods (eg breath test labeled isotopes, ELISA and immunoblot methods) which are complex, time consuming and high cost.

International guidelines suggest detection of IgG antibodies to confirm the infection of Helicobacter pylori (Chey & Wong, 2007˙Malfertheiner et al, 2007). In uncommon cases, however, as acute infection, the detection of IgM antibodies may be useful and supportive (She et al, 2009). Further studies showed that the presence of IgA antibodies detect cases of infection with Helicobacter pylori in which IgG are negative (Urita et al, 2004). Therefore, the addition of the IgA isotype is appropriate to include the rapid detection assays of Helicobacter pylori, to be used by a specialist in cases that require increased investigative.

DyonHpAb®triple (*H. pylori* Ab IgG, IgM, IgA) is a rapid, non-invasive, highly accurate and easy to perform qualitative test that allows a health professional to detect antibodies to human IgG, IgM, IgA against Helicobacter pylori in samples of whole blood, capillary blood, serum or plasma.

PRINCIPLE OF THE METHOD

DyonHpAb®triple is a qualitative immunoassay of the sandwich-type colloidal gold to detect antibodies IgG, IgM, IgA against Helicobacter pylori. The method utilizes a membrane coated with antibodies in order to detect antibodies against H. pylori selectively and with great sensitivity. At the start of the test, the sample is mixed with the complex of the chromophore and begins to move through the membrane. If the sample contains antibody, it will interact with the pre-capped complex of chromophore, so you'll see a color (purple) line in the Test zone (T). Both negative and positive samples should generate a colored line (purple) in the Control Zone (C), which is indicated the proper execution of technique, sample volume and functioning of examination.

STORAGE AND STABILITY

Store the kit at 2-30 ° C. If the kit was stored in the refrigerator, it should be transferred to room temperature before proceeding with the examination. Under these conditions, the kit is stable until the expiration date printed on the packaging.

PACKAGE CONTENTS

Each kit contains 1 individually wrapped (or multiples thereof), as follows:
-Pin-scarification
-Vial with Dilution
-Sealed plastic package with test cassette
-1 package insert

SAMPLE COLLECTION AND PREPARATION

A. Collection of Whole Blood, Serum or Plasma: Venous whole blood collected by conventional methods in the presence of anticoagulant blood collection should be tested within 24 hours of collection. In between, the sample can be stored in the refrigerator. Serum or plasma should be separated immediately to avoid hemolysis; the resulting samples should be tested immediately after separation. Serum and plasma can be stored in the refrigerator up to 3 days, while in long term, they should be stored in the freezer (-20 ° C).

B. Capillary Blood Collection.

1. Remove the yellow strain of the pin scarification to unlock it.
2. Disinfect the fingertip of the patient with alcohol. Knead the fingertip to increase blood circulation.
3. Apply disengaged pin on the finger. In this position there is no contact of the pin with the finger.
4. Press decisively the yellow button on the peak of the pin.

5. After scarification knead the pierced point to display one large drop of blood. The capillary blood is to be used immediately.

TEST PROCEDURE-INTERPRETATION OF RESULTS

Before the examination, make sure that the plastic packaging with test cassette has come to room temperature. The test cassette should unsealed only shortly before the examination.

6. Turn the pierced finger of patient and press it that 1 drop (25ml) of serum or plasma or 1 drop of capillary (or whole) blood fall into the special hole round cartridge examination.

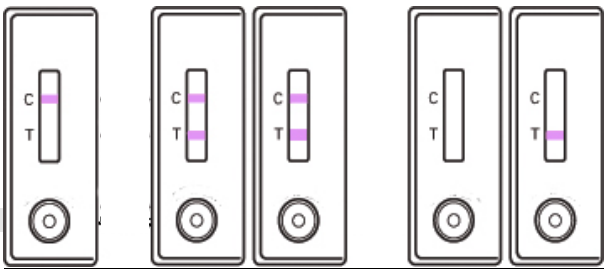
7. Immediately open the Dilution Bottle and pour 3 drops of Dilution Solution in the special round hole on the cartridge.

8. Start the timer (not supplied).

9. Note the solubilized sample as a red-purple front which moves to zones (T) and (C). Interpret the results in 10 minutes. Do not interpret after 15 minutes.

Positive results = 2 color lines.
The test is positive for antibodies to Helicobacter pylori if two colored lines appear. A color purple line will appear in the Test Zone (T) and one purple in the Control Zone (C). This fact indicates the presence of antibodies to Helicobacter pylori in the patient's blood. Any color line in Zone (T) corresponds to a positive result. The intensity of color in the Zone (T) may be stronger or paler of color intensity in Zone (C).

Negative Positive Invalid



Negative Results = 1 color line
The test is negative if only (1) colored line appears in the Control Zone (C). The interpretation of a negative result is guaranteed in 15 minutes.

Invalid = Absence of colored line in the Control Zone (C)
The test is invalid if after 15 minutes there is no line appeared in the Control Zone (C), even if a colored line has appeared in the Test Zone (T). If this happens reread the instructions and repeat the test using a new device. If the problem persists, contact Dyonmed S.A.

PRECAUTIONS

Take precautions regarding the collection, handling, storage and disposal of waste samples and kit components. Handle all specimens as if they contain infectious agents. Take precautions against microbiological hazards throughout the procedure. Apply standard procedures for proper disposal of specimens. Operation is not influenced by known environmental factors other than temperature.

DyonMed S.A.
Point of Care Solutions

182-184 Lavriou Av., 15354, Peania, Attika
Tel. (+30) 210.66.55.494 & -495, Fax: (+30) 211.770.8599
info@dyonmedical.com www.dyonmedical.com
ISO 9001:2008 (No. 010110003)| ISO 13485:2003 (No 01210005)
by TÜV Austria Hellas

RESTRICTIONS

1. The test results of DyonHpAb®triple must be integrated with other symptoms, clinical findings or diagnostic results in order to be a diagnosis.
2. No rapid test can exclude the presence of bacteria, especially at the beginning of infection.
3. The test does not differentiate between IgG, IgM or IgA. Accordingly, at least one of the above classes of antibodies is present on the positive results.
4. Negative results that are inconsistent with symptoms of peptic discomfort should drive in retesting or further testing.
5. Positive results may be due to active infection or latent bacteria, so not mandatory indicate gastrointestinal diseases.
6. The characteristics are valid only if the contents of the package are in good condition.
7. **DyonHpAb®triple** device is for single use only. If a dissolved sample should be retested use a new device.
8. The test **DyonHpAb®triple** addressed exclusively to healthcare professionals.

QUALITY CONTROL

Internal Quality Control: The colored line appears in the Zone (C) is an internal control procedure. It assures proper sample volume and proper technical procedure. A clean background is the internal negative control. If the test works correctly, the background of Control Zone should be white to light pink and it should not be complicates the result interpretation.

External Quality Control: Each laboratory using the test should develop its own test instructions and process certification of the test.

OPERATING CHARACTERISTICS

1. Performance. The test **DyonHpAb®triple** tested for detection of infection with H. pylori in clinical study with 310 patients aged 17-77 years, of whom 62% were male and 38% female. **DyonHpAb®triple** compared with biopsy and histological examination (golden standard). At first all samples were identified as positive or negative by histological examination. Then the same samples were tested with **DyonHpAb®triple** in a blinded manner (the experimenter did not know which samples were negative or positive when performing the tests). Under these conditions the performance of device **DyonHpAb®triple** was:

- Specificity and sensitivity of the device **DyonHpAb®triple** compared with biopsy and histological examination: 93.3% and 93.8% respectively *

- Positive Predictive Value of the device **DyonHpAb®triple** compared to biopsy and histological examination: 93.3%*

- Negative Predictive Value of the device **DyonHpAb®triple** compared with biopsy and histological examination: 93.8%*

- Accuracy of the device **DyonHpAb®triple** compared with biopsy and histological examination: 93.6%*

* regards confidence interval CI=95%

2. Specificity. Blood samples from patients positive or negative in H. pylori infected with *Campylobacter fetus*, *Campylobacter jejuni* or *E. coli* and analyzed by the device **DyonHpAb®triple** in 10 iterations at a time. Without exception the samples gave the expected results, i.e. were positive or negative regardless of the additional infecting bacterium.

3. Cross-Reaction. Blood samples from negative patients to H. pylori which had been identified by histological examination were transfected with the following infecting agents and analyzed by the device **DyonHpAb®triple** in 5 iterations at a time. Without exception the samples gave the expected results, i.e. negative regardless of interpolating factor.

acetaminophen, 20 mg/dl	glucose, 2000 mg/dl
aspirin, 20 mg/dl	hemoglobin 500 mg/dl
ascorbic acid, 20 mg/dl	ketone, 40 mg/dl
atropine, 20 mg/dl	mestranol 3 mg/dl
bilirubin, 60 mg/dl	nitrates, 20 mg/dl
caffein, 20 mg/dl	penicillin, 40.000 U/dl
creatinine, 20 mg/dl	Sodium Heparin, 3 mg/dl
gentisic acid, 20 mg/dl	Lithium Heparin, 3 mg/dl

4. Unlike other methods for detecting H. Pylori, the test **DyonHpAb®triple** is not affected by simultaneous administration of proton pump inhibitors or any of the patient's diet. This is due to the fact that the assay **DyonHpAb®triple** is based on precipitation of the microbe antigen by the specific antibody to this. The antibody reacts with the antigen through multiple weak electrostatic bonds and forces van der Waals, which ensure high chemical affinity binding regardless of where the antigen is from.

REFERENCES

Atherton JC and Blaser MJ (2009). Coadaptation of Helicobacter pylori and humans: ancient history, modern implications. J. Clin. Invest. 119: 2475–2487.

Chey WD, Wong BC (2007) Practice Parameters Committee of the American College of Gastroenterology. **American College of Gastroenterology guideline on the management of Helicobacter pylori infection**. Am J Gastroenterol. 2007 Aug;102(8):1808-25. Epub 2007 Jun 29.

Dorer MS, Talarico S and Salama NR (2009), Helicobacter pylori 's Unconventional Role in Health and Disease. PLoS Pathogens, Volume 5 issue 10 | e1000544.

Feinstein LB, Holman RC, Yorita Christensen KL, Steiner CA. Swerdlow, DL (2010). Trends in Hospitalizations for Peptic Ulcer Disease, United States, 1998–2005. Emerging Infectious Diseases • www.cdc.gov/eid • Vol. 16, No. 9

Malfertheiner P, Megraud F, O'Morain C, Bazzoli F, El-Omar E, Graham D, Hunt R, Rokkas T, Vakil N, Kuipers EJ, The European Helicobacter Study Group (EHSG) (2007), Current concepts in the management of Helicobacter pylori infection: **the Maastricht III Consensus Report**. Gut 56:772–781.

She RC, Wilson AR, Litwin CM. (2009) Evaluation of Helicobacter pylori Immunoglobulin G (IgG), IgA, and IgM serologic testing compared to stool antigen testing. Clin Vaccine Immunol. Aug;16(8):1253-5. Epub 2009 Jun 10.

Urita Y, Hike K, Torii N, Kikuchi Y, Kurakata H, Kanda E, Sasajima M, Miki K. (2004). Comparison of serum IgA and IgG antibodies for detecting Helicobacter pylori infection. Intern Med. Jul;43(7):548-52.

24-hour Help Line for Medical Devices
Tel. (+30) 211.800.8167
Strictly professional in vitro diagnostic use

DyonHpAb®triple (IgG, IgM, IgA)
Genuine Product **Point of Care®**
Produced in U.S.A. on behalf of DyonMed S.A.