Advantage

LEPTOSPIRA IgM & IgG CARD

Rapid Diagnostic Test For the Qualitative Detection of Leptospira Specific IgM and IgG antibodies in Human Serum/Plasma

INTENDED USE

Advantage Leptospira IgM & IgG Card is a visual, rapid, sensitive, qualitative immnunoassay for the detection of Leptospira specific IgM and IgG antibodies in human serum or plasma.

INTRODUCTION

Leptospirosis is a world-wide occuring zoonotic disease, caused by infection with pathogenic spirochetes of the genus Leptospira. Although traditionally considered as an occupational risk among persons exposed to contaminated water or infected animal urine, leptospirosis is becoming recognised as a common cause of febrile illness in tropical environmments world-wide. The organism enters the human body through cuts or abrasions on the skin or through intact mucosa of the mouth, nose or conjunctiva. The clinical manifestations of leptospirosis range from a mild catarrh like fever, chills, nausea, muscle aches to icteric disease such as Weil's syndrome, which are charaterized by renal failure, liver impairments & haemorrhages and have a high mortality rate. As clinical symptoms & signs of this infection resemble those of many other infectious diseases including Viral haemorrhage fever and Dengue fever, clinical findings need to be confirmed by laboratory diagnostic techniques.

PRINCIPLE (ANTIGEN-ANTIBODY REACTION)

Advantage Leptospira IgM & IgG Card is based on immunochromatographic assay principle. The test uses monoclonal anti-human IgM antibody (test line M) and monoclonal anti-human IgG (test line G) immobilised on a nitrocellulose strip. The test sample is introduced to and flows laterally through an absorbent pad where it mixes with the conjugate. The conjugate contains colloidal gold conjugated to leptospira antigen which is prepared from culture of micro-organisms. If the sample contains Leptospira specific IgM and/or IgG antibodies, it forms a complex with the leptospira antigen gold conjugate making antigen antibodies complex. This complex then migrates through the nitrocellulose membrane by caplillary action. When the complex meets the line of immobilized monoclonal antibodies (Test line, 'M & G'), it generates a red line, indicating that the sample is reactive. A red procedural control line should always develop at 'C' region to indicate that the test has been performed properly.

MATERIAL PROVIDED

1. Advantage Leptospira IgM & IgG Card
2. Sample Dropper
3. Instruction Manual

DESCRIPTION OF SYMBOLS USED

The following are graphical symbols used in or found on J. Mitra diagnostic products and packing. These symbols are the most common ones appearing on medical devices and their packing. They are explained in more detail in the European Standard EN ISO 15223-1:2016.

	Manufactured By	IVD	*In vitro* diagnostic medical device
Σ	No. of tests		See Instruction for use
LOT	Lot Number Batch Number	30°C 2°C	Temperature Limitation
	Manufacturing Date		Caution, see instruction for use
	Expiry Date	REF	Catalogue Number
	Do not use if package is damaged	EC REP	Authorized Representative in the European Community
	Keep away from sunlight	2	Single Time use only

KIT PRESENTATION

10 Test Pack

KIT STORAGE & STABILITY

The kit should be stored at 2-30°C in the coolest and driest area available. Expiry date on the kit indicates the date beyond which kit should not be used. The kit should not be frozen and must be protected from exposure to humidity. Advantage Leptospira IgM & IgG Card should be used within one hour after removal from the foil pouch.

WARNING FOR USERS

CAUTION: ALL THE SAMPLES TO BE TESTED SHOULD BE HANDLED AS THOUGH CAPABLE OF TRANSMITTING INFECTION. NO TEST METHOD CAN OFFER COMPLETE ASSURANCE THAT HUMAN BLOOD PRODUCTS WILL NOT TRANSMIT INFECTION.

1. The use of disposable gloves and proper biohazardous clothing is STRONGLY RECOMMENDED while running the test.
2. In case there is a cut or wound in hand, DO NOT PERFORM THE TEST.
3. Do not smoke, drink or eat in areas where specimens or kit reagents are being handled.
4. Tests are for *in vitro* diagnostic use only and should be run by competent person only.
5. Do not pipette by mouth.
6. All materials used in the test and samples should be disposed off in accordance with established safety procedures/ guidelines.
7. Wash hands thoroughly with soap or any suitable detergent, after the use of the kit. Consult a physician immediately in case of accident or contact with eyes, in the event that contaminated material are ingested or come in contact with skin puncture or wounds.
8. Spills should be decontaminated promptly with Sodium Hypochlorite or any other suitable disinfectant.
9. Do not open the foil pouch to remove the product until it attains room temperature and you are ready to perform the test.
10. **Optimal test performance requires strict adherence to the test procedure described in the instuction manual.**

PRECAUTIONS

1. Do not reuse test card.
2. Do not use kit beyond the stated expiry date mentioned on the kit.
3. Interpret the results at the end of 20 minutes only.
4. Use a separate sample dropper for each sample and discard it as biohazardous waste.

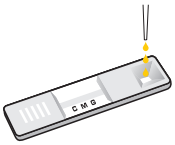
SPECIMEN COLLECTION & STORAGE

1. Advantage Leptospira IgM & IgG Card test should be performed in human serum or plasma only immediately after collection.
2. If not tested immediately, specimen should be refrigerated at 2-8°C for upto 24 hours following collection.
3. If testing within 24 hours is not possible, specimen should be frozen at -20°C for 3 months or -70°C for longer period.
4. Specimen containing visible precipitates or cloudy specimens may give inconsistent test results. Such specimens should be clarified prior to testing by high speed centrifugation i.e. 10,000 rpm for 15 minutes before testing.
5. Haemolysed specimen or specimen with microbial contamination should be discarded and fresh aliquot should be collected.
6. Repeated freezing & thawing of the specimen should be avoided.

TEST PROCEDURE

R.T. 20-30°

1. Bring the complete kit & sample to be tested to room temperature prior to testing. Once the test kit is opened, it should be used within one hour.
2. Remove the test card from the foil pouch prior to use label the test card with patient name or identitification number.
3. Add 3 drops (100 μl) of the sample using sample dropper into the sample well of Leptocard. Dispose off the dropper considering it to be Biohazardous.



5. Allow reaction to occur during the next 20 minutes.
6. Read results at 20 minutes. Positive results may appear as early as 2-10 minutes. However, negative results must be confirmed at 20 minutes only.
7. Discard the Advantage Leptospira IgM & IgG Card immediately after reading result at 20 minutes, considering it to be potentially infectious.

Important: Do not read results after 20 minutes as it may give erratic results.

INTERPRETATION OF RESULT

IgM & IgG REACTIVE

Fig. a

As shown in Fig. a, appearance of red coloured line in the control region 'C' and Test region; IgM region 'M' and IgG region 'G' indicates that the sample is reactive for both leptospira IgM & IgG antibodies.

IgM REACTIVE

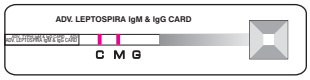


Fig. b

As shown in Fig. b, appearance of red coloured line in the control region 'C' and Test region; IgM region 'M' indicates that the sample is reactive for leptospira IgM antibodies.

IgG REACTIVE

Fig. c

As shown in Fig. c, appearance of red coloured line in the control region 'C' and Test region; IgG region 'G' indicates that the sample is reactive for leptospira IgG antibodies.

NON-REACTIVE

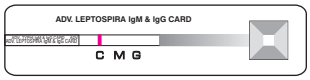


Fig. d

As shown in Fig. d, appearance of one distinct red coloured line in the control region 'C' only (with no line in the IgM region 'M' & IgG region 'G') indicates that the sample is non-reactive for leptospira antibodies.

INVALID

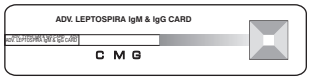


Fig. e

Fig. f

Fig. g

The test is invalid if no control line appear after completion of test either with clear background or pinkish background as shown in (Fig. e, f & g). The test should be treated as Invalid which may be because of following reasons:

(a) Improper storage at temperature other than the recommended temperature.

(b) Wrong Procedure

(c) Long atmospheric exprosure of the test device after opening the pouch.

The test should be repeated using a new device and test sample.

LIMITATIONS OF THE PROCEDURE

1. The Advantage Leptospira IgM & IgG Card is for *in vitro* diagnostic use only.
2. The test is a qualitative screening assay and is not suggested for use in determining quantitative levels.
3. This is only a screening test. All samples detected reactive must be confirmed by using confirmatory test such as MAT (Microscopic Agglutination Test). Therefore for definitive diagnosis, the patient's clinical history, symptomatology as well as serological data, should be considered.
4. If Advantage Leptospira IgM & IgG Card test is non-reactive and clinical symptoms persist, the test should be repeated with a second sample collected at a later date.
5. False positive results can be obtained due to cross reaction with Epstein-BARR virus, Influenza A & B, Brucella, Dengue Virus. This occurs in less then 1% of the sample tested.

PERFORMANCE CHARACTERISTICS

An elaborated study has been done in-house on Advantage Leptospira IgM & IgG Card to determine its performance as a screening test. The performance of the test was evaluated and compared with a licensed commercially available ELISA test kit in-house by using a known panel of Serum/ Plasma Lepto negative & positive samples. The samples included cross-reacting samples; Epstein-Barr virus, Malaria, Rheumatoid factor, Japanese encephalitis, yellow fever and West Nile viruses. The results obtained are as follows:

Sample Type	No. of Samples tested	Result of licensed test	Adv. Leptospira IgM & IgG Card results
Negative for Ab to Leptospira	2590	2589	2585
Leptospira IgM Positive	108	108	107
Leptospira IgG Positive	10	10	10

Sensitivity : 98.16% (for IgM Antibodies) & 100% (for IgG Antibodies)

Specificity : 99.61%

Precision: Within run (Intra assay) & between run (Interassay) precision have been determined by testing 10 replicates of five specimens - one negative, three Lepto IgM positive and one Lepto IgG Positive. The C.V. (%) of all the five samples were within 10% of the time.

LIMITED EXPRESSED WARRANTY DISCLAIMER

The manufacturer limits the warranty to the test kit, as much as that the test kit will function as an in-vitro diagnostic assay within the limitations and specifications as described in the product instruction-manual, when used strictly in accordance with the instructions contained therein. The manufacturer disclaims any warranty expressed or implied including such expressed or implied warranty with respect to merchantability, fitness for use or implied utility for any purpose. The manufacturer's liability is limited to either replacement of the product or refund of the purchase price of the product and in no case liable to for claim of any kind for an amount greater than the purchase price of the goods in respect of which damages are likely to be claimed. The manufacturer shall not be liable to the purchaser or third parties for any injury, damage or economic loss, howsoever caused by the product in the use or in the application there of.

BIBLIOGRAPHY

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For *in-vitro* diagnostic use only, not for medicinal use

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