

# DENGUE NS1 and IgG+IgM Test Kit



For Professional Use

A Rapid Card Test to detect **Dengue NS1 Antigen** & differential detection of **IgG and IgM antibodies** in human Serum or Plasma



Read the pack insert before use provided along with the kit

REF DgmNS1

**INTENDED USE:** Dengue is a flavivirus found largely in areas of the tropic and sub tropics. There are four distinct but antigenically related serotypes of dengue viruses, and transmission is by mosquito, principally *Aedes aegypti* and *Aedes albopictus*. The mosquito-borne dengue viruses (serotypes 1-4) cause dengue fever, a severe flu like illness. The disease is prevalent in third world tropical regions and spreading to sub tropical developed countries-including occur worldwide each year, including potentially deadly form of the disease called dengue haemorrhagic fever (DHF) and dengue shock syndrome (DSS). Primary infection with dengue virus results in a self-limiting disease characterized by mild to high fever lasting for 3 to 7 days, severe headache with pain behind the eyes, muscle and joint pain, rash and vomiting. Secondary infection is the more common form of the disease in many parts of Southeast Asia and South America. IgM antibodies are not detectable until 5-10 days in case of primary dengue infection and until 4-5 days in secondary infection after the onset of illness. IgG appear after 14 days and persist for life in case of primary infection and rise within 1-2 days after the onset of symptoms in secondary infection. This form of the disease is more serious and result in DHF and DSS. The major clinical symptoms can include high fever, haemorrhagic events, and circulatory failure, and the fatality rate can be high as 40%. Early diagnosis of DSS is particularly important, as patient may die within 12 to 24 hours if appropriate treatment is not administered. Primary dengue virus infection is characterized by elevation in specific NS1 antigen levels 0 to 9 days after the onset of symptoms; this generally persists upto 15 days. Earlier diagnosis of Dengue reduces risk of complication such as dengue haemorrhagic fever (DHF) and dengue shock syndrome (DSS), especially in countries where dengue is endemic.

## TEST PRINCIPLE:

**DENGUE NS1 Antigen test:** The Bhat Bio-Scan® Dengue NS1 Antigen Card test contains a membrane strip which is precoated with antibodies to Anti Dengue NS1 antigen. This test device has a letter of T & C as "Test line" and "Control line" on the surface of the case. Both the Test Line (T) and Control line (C) in result window are not visible before applying any samples. When a sample is added to the device, Dengue NS1 antigen if present in the sample will bind to the anti-dengue NS1 gold colloid conjugate making antigen antibodies complex. This complex migrates along the membrane to the test region and forms the visible pink line at "T" as antibody-antigen. The Control Line is used for Procedural control. Control Line should always appear if the test procedure is performed properly and reagents of control line are working.

**DENGUE IgG+IgM CARD TEST:** The Dengue IgG/IgM test kit is a sandwich immuno chromatographic membrane based screening test to detect the antibodies for dengue virus. The test can be used with Serum or plasma. The test employs the use of colloidal gold particle conjugated with IgG & IgM binding proteins and a unique combination of dengue antigen immobilized on the membrane at T. After the addition of sample and running buffer in the sample well of test device, the mixture passes through the conjugate gold and makes an immune complex.

The immune complex will bind with the antigen immobilized at test zone (IgG & IgM) of the test device if antibodies to dengue present in the sample. The remaining complex continues to migrate to a control area (C) in the test device and produces a colored band in the control (C) zone. The control band indicates that the test has been performed properly and is in working condition. Appearance of pink/purple band at test (IgG & IgM) in addition to band at control (C) indicates that sample is positive for dengue antibodies.

**STORAGE AND STABILITY :** Kit should be stored between 2-30°C in the sealed pouch. The kit is stable until the expiry date mentioned on the pouch when stored under the above conditions.

PACK SIZE: 10 Test

## CONTENTS OF THE KIT

Dengue NS1 Ag & IgG+IgM Test Devices	10 Nos.
Silicagel	10 Nos.
Test Diluent	1.4 ml
5µl Dropper	10 Nos.
50µl Dropper	10 Nos.
Pack Insert	1 No.

## MATERIAL REQUIRED BUT NOT PROVIDED:

- Sterilized vial
- Disposable gloves
- Micro pipette (1-10µl)
- Sodium hypochlorite solution (free available chlorine 50-500mg/L)
- Autoclaved Tips

## PRECAUTIONS :

- Read this Pack insert carefully.
- DO NOT FREEZE THE KITS.
- Do not use after the expiration date.
- Use only serum or plasma specimen. Furthermore, do not use umbilical cord blood because it prevents colloidal gold from migration and can interfere with results.
- Carefully observe the prescribed number of drops if be added 2 drops (100µl) of Serum or Plasma only for Dengue NS1 Antigen Test and 5ul of Serum or plasma and 2 drops of diluent Dengue IgG+IgM Test.
- The Dengue NS1 and IgG+IgM Test kit pack if refrigerated should be brought down to room temperature before testing. Assay should be conducted at room temperature (15- 30°C)
- Use the kit soon after removing from the pouch.
- Do not use the test device, if the pouch seal is broken.
- Frozen specimens should be brought down to room temperature before testing.
- Specimen should not be repeatedly frozen and thawed.
- For in vitro diagnostic use only.

All human serum and plasma samples should be considered potentially infectious. It is recommended that all specimens of human origin should be handled as recommended for any potentially infectious human serum or blood specimen in the Centers for Disease Control / National Institute of Health manual "Biosafety in Microbiological and Biomedical Laboratories", 1984. Never pipette by mouth. Do not smoke, eat or drink in areas in which specimens or kit reagents are handled. Afterwards wash hands carefully. Avoid splashing or forming aerosols. Discard all materials and specimens as if capable of transmitting infection. The preferred method of disposal is autoclaving for a minimum of one hour at 121°C. Liquid waste not containing acid may be mixed with sodium hypochlorite so that the final mixture contains 50-500 mg/l available chlorine. Allow 30 minutes for decontamination.

**SPECIMEN :** Fresh Serum or plasma.

## Specimen collection & Preparations:

- Collect blood in a clean, dry, sterilized vial and allow it to clot. Separate the serum by centrifugation at 5000 r.p.m for 15 minutes at room temperature.
- If serum is not to be assayed immediately it should be stored at 2-8°C or if storing more than 3 days, then freeze the specimen at -20°C or below.

**ASSAY PROCEDURE**

**A. For NS1 Antigen Test**

1. Bring the pouch to room temperature.
2. Remove the device from the pouch just prior to testing.
3. Place the device on a flat surface.
4. Add 2 drops (100 µl) of sample into the sample window (S)
5. Read the results in 20 minutes. **DO NOT READ ANY RESULT BEYOND 20 Minutes**
6. Any line appearing after Twenty minutes would be of no diagnostics value.

**B. For IgG + IgM Test**

1. Bring the pouch to room temperature.
2. Remove the device from the pouch just prior to testing.
3. Place the device on a flat surface.
4. Add 5µl (Fill the sample up to the mark in the dropper provided) of serum or plasma into the sample window(S)
5. Add 2 drops of diluent provided in the dropper bottle into the same sample window (S)
6. Read the results in 20 minutes. **DO NOT READ ANY RESULT BEYOND 20 Minutes**
7. Any line appearing after Twenty minutes would be of no diagnostics value.

**INTERPRETATION OF RESULTS**

**A. Dengue NS1 Antigen test**

**1. NEGATIVE**

If only one red line (Control Line) appears in the result area, Interpret the result as negative. This shows that the specimen does not contain Dengue NS1 antigen. (Fig. A)



Fig. A DENGUE NS1 NEGATIVE

**2. POSITIVE**

If two red lines (Control & Test) appear in the result area the specimen is reactive for Dengue NS1 antigen.(Fig.B).



Fig. B DENGUE NS1 POSITIVE

**3. INVALID**

If no line appears after the test is complete, interpret the result as invalid. This shows that the test has been performed incorrectly or there was some procedural error. Please check the procedure and retest using a new device.(Fig.C)



Fig. C DENGUE NS1 IN VALID

**B. Dengue IgG+IgM Test**

**1. NEGATIVE**

If only one red line (Control Line) appears in the result area, Interpret the result as negative. This shows that the specimen does not contain Dengue IgG and IgM antibodies. (Fig. 1)



Fig. 1 DENGUE Ab NEGATIVE

**2. POSITIVE**

If two red lines (Control & IgG) appear in the result area the specimen is reactive for Dengue IgG antibodies.(Fig.2).

If two red lines (Control & IgM) appear in the result area the specimen is reactive for Dengue IgM antibodies.(Fig.3).

If three red lines( Control IgG and IgM) appear in the result area the specimen is reactive for Dengue IgM and IgG antibodies.(Fig.4).



Fig. 2 DENGUE IgG POSITIVE



Fig. 3 DENGUE IgM POSITIVE



Fig. 4 DENGUE IgG+IgM POSITIVE

**3. INVALID**

If no line appears after the test is complete, interpret the result as invalid. This shows that the test has been performed incorrectly or there was some procedural error. Please check the procedure and retest using a new device.(Fig.5)

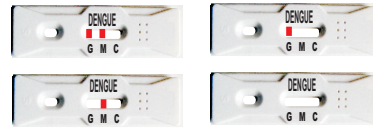


Fig. 5 DENGUE Ab IN-INVALID

**IMPORTANT**

Test line either dark or light in color (red) should be considered reactive for Dengue NS1 antigen and Dengue IgG and IgM antibodies.

**Expected Value** : Primary dengue is characterized by the presence of detectable IgM 3-5 days after the onset of infection. Secondary dengue is characterized by the elevation of specific IgG 1- 2 days after the onset of infection and in the majority of cases this is generally accompanied by an elevation of IgM.

**Sensitivity**: 98.4% for IgG+IgM Test & 100% for NS1 Antigen Test

**Specificity**: 98.4% for IgG+IgM Test & 99.5% for NS1 Antigen Test

**LIMITATIONS OF THE TEST**

This test detects the presence of antibodies to dengue in the Specimen and should not be used as the sole criteria for the diagnosis of Dengue virus infection.

1. In early infections and some secondary infections, detectable levels of IgM antibodies may be low.
2. Some patients may not produce detectable levels of antibody within the first 7-10 days after infection. Where symptoms persist, patients should be retested 3-4 days after the first specimen.
3. If the test result is negative and clinical symptoms persists, additional follow-up testing using other clinical methods is recommended. A negative result does not preclude the possibility of an early infection of Dengue virus.

**NOTE::** Even after the best effort is made to supply the product as per the sample submitted but due to continuous R & D, the company reserves the right to improve/change any specifications/components without prior information/notice to the buyer

**LIMITED EXPRESSED WARRANTY OF MANUFACTURER**

The manufacturer limits the warranty to this test kit, as much as that the test kit will function as an in vitro diagnostic assay within the Nature of Sample, Procedure limitations and specifications as described in the product instruction manual, when used strictly in accordance with the instructions contained. 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 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 thereof.

**REFERENCES :**

1. Dengue haemorrhagic fever : Diagnosis, treatment, prevention and control. WHO 2nd Edition 1997.
2. Songee L. ranch and Paul N. Levett. Evaluation of four methods for detection of immunoglobulin M antibodies to dengue virus. Clin.Diagn. Lab. Immunol. Vol6 (4) p 555-557, 1999.
3. Lamm S.K. (1995). dengue haemorrhagic fever. Rev. Med. Micro,6-39-48.
4. Seth, J. (1991). standardization & quality assurance. In principle and practice of immunoassay, Ed. C.P. Price & D.J. Newman. Macmillan Publishers, pp.154-189.

BS ISO-15223-1:2012(E) MEDICAL DEVICES SYMBOL					
	Temperature Limitation 2°C		Date of Manufacture 2001-06		In vitro Diagnostic Device
	Batch Code		Company name & address		Consult Instructions For Use
	Use by		Company Name		Authorised Representative in European Community
	Do Not Reuse		Sufficient for		KEEP AWAY FROM SUNLIGHT
	KEEP DRY		NON-STERILE		NEGATIVE CONTROL
	POSITIVE CONTROL				

