

Size : 137 x 218 mm

Virdict4

DEVICE

Rapid Immunochromatographic Test System Pack for Simultaneous Detection of HIV 1/2 antibodies, HCV antibodies, Syphilis antibodies and HBsAg in human Serum/Plasma

INTENDED USE

Virdict 4 is a Rapid Immunochromatographic test for simultaneous detection of HBsAg and antibodies to HIV, HCV and Syphilis in human serum/plasma.

SUMMARY

Acquired immune deficiency syndrome (AIDS) caused by at least two types of retrovirus HIV 1 and HIV 2, collectively referred as HIV1/2. Antibody to HIV 1 core protein p24, transmembrane protein gp 41 and/or antibodies to HIV 2 transmembrane protein gp 36 are present in sera of individual infected with AIDS. HIV in **Virdict 4** detects these antibodies which indicates exposure to the HIV 1/2 or both viruses.

HCV is a small, enveloped single stranded RNA virus containing a linear genome with a length of about 9,600 nucleotide having positive polarity. It is the major cause of permanently transmitted non-A, non-B hepatitis. Antibodies to HCV are reported in 80% on non-A, non-B hepatitis patients. HCV infection frequently progresses to chronic liver disease. Based on phylogenetic analysis, HCV has been grouped into 6 major genotype each of which contains one or more subtype. HCV in **Virdict 4** detects anti – HCV antibodies ensuring detection of all antibody isotypes viz. IgM, IgG, IgA etc. for all 6 genotypes.

Syphilis is a sexually transmitted (venereal) disease caused by the spirochete *Treponema pallidum*. The disease can also be transmitted congenitally thereby attaining its importance in antenatal screening. After infection the host forms non-treponemal anti lipoidal antibodies to the lipoidal material released from the damaged host cells as well as *Treponema* specific antibodies. Serological tests for non-treponemal antibodies such as VDRL, RPR, and TRUST etc. are useful as screening tests. Tests for *Treponema* specific antibodies such as TPHA, FTA-ABS, rapid *Treponema* antibody tests are gaining importance as screening as well as confirmatory tests because they detect the presence of antibodies specific to *Treponema pallidum*. Syphilis in **Virdict 4** is modified TPHA, which qualitatively detects the presence of IgM and IgG class of *Treponema* specific antibodies during syphilis infection.

Blood containing hepatitis B virus (HBV) is potentially infectious. Hepatitis B surface antigen (HBsAg), earlier known as Australian antigen is among the first serological markers that circulate in blood of infected person even two to three weeks prior to appearance of clinical symptoms. The level of HBsAg is especially elevated during the symptomatic phase. Detection of HBV using HBsAg as marker to screen blood donors is essential to reduce the risk of HBV transmission through blood transfusion. HBsAg detection is also useful for screening high risk group for HBV and for differential diagnosis of Hepatitis infection. HBsAg in **Virdict 4** detects presence of HBsAg in serum/plasma specimen qualitatively.


PRINCIPLE

The device utilizes the principle of immunochromatography immunoassay on a membrane. As the test sample flows through the membrane assembly of the test device, the colored HIV specific recombinant antigen-colloidal gold conjugate, recombinant *Treponema pallidum* antigen- colloidal gold conjugate, a multi – epitope HCV recombinant peptide antigen conjugated to colloidal gold and a colored anti HBsAg colloidal gold conjugate bind to the specific antibodies and antigen. The respective complexes move further to the test regions and get immobilized to form test band/s.

The unreacted conjugate and unbound complex, if any along with rabbit IgG conjugate move further on the membrane. Subsequently rabbit IgG conjugate gets immobilized by the goat anti – rabbit antibodies coated on the membrane at the control region (C), forming a pink to pink-purple colored band. This control band serves as a procedural control and thus aids to validate the results.

REAGENTS AND MATERIALS SUPPLIED

- A. The kit contains individual pouches each containing a
1. **DEVICE** Test Device
 2. **PIPETTE** Sampler Applicator
 3. Desiccant pouch
- B. **BUF** Diluent buffer
- C. Package insert.

REF	402090025	402090050
	25 T	50 T

OPTIONAL MATERIAL REQUIRED

Stopwatch.

STORAGE AND STABILITY

The sealed pouches in the test kit may be stored between 4 - 30°C till the duration of the shelf life as indicated on the pouch. DO NOT FREEZE.

NOTE

1. For in vitro diagnostic and professional use only. NOT FOR MEDICINAL USE.
2. Do not use beyond expiry date
3. Read the instructions carefully before performing the test.
4. Handle all specimen as if potentially infectious.
5. Follow standard biosafety guidelines for handling and disposal of potentially infectious material.
6. If desiccant colour at the point of opening the pouch has turned from blue to pink or colourless, another test device must be run.

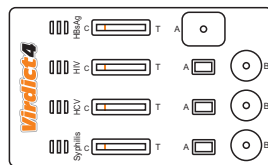
SPECIMEN COLLECTION AND PREPARATION

1. No prior preparation of the patient is required before sample collection by approved techniques.
2. Fresh serum / plasma is preferable. Serum / plasma may be stored at 2-8 °C up to 24 hours in case of delay in testing. For long term storage, freeze the specimen at -20 °C.
3. Repeated freezing and thawing of the specimen should be avoided.
4. Do not use clotted, contaminated, viscous/turbid specimen.
5. Specimen containing precipitates or particulate matter must be centrifuged and the clear supernatant only used for testing.
6. Frozen samples for retrospective studies must be centrifuged at 3000 rpm for 15 minutes and the clear supernatant must be used for tests.

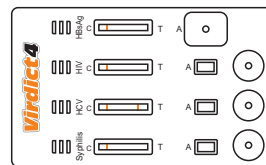
TEST PROCEDURE

1. Bring the sealed aluminium foil pouch of Virdict 4 membrane test assembly to room temperature.
2. Open a foil pouch by tearing along the "notch".
3. Remove the membrane test assembly. Once opened, the membrane test assembly must be used immediately.
4. Label the membrane test assembly with specimen identity.
5. Place the membrane test assembly on a flat horizontal surface.
6. Using the dropper provided carefully dispense exactly **two drops** of serum or plasma into the sample port "A" of HBsAg and **One drop** of serum/plasma in to the sample port "A" of HIV, HCV and Syphilis respectively.
7. Add **three drops** of diluent buffer into the port "B" of HIV, HCV and Syphilis respectively.
8. Read the results at the end of 30 minutes.

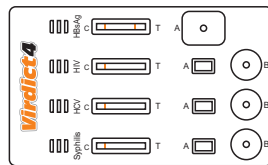
INTERPRETATION OF RESULTS



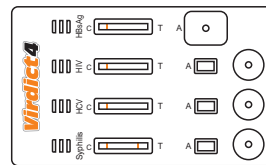
Negative for HBsAg, HIV, HCV & Syphilis



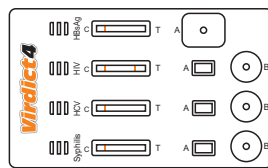
Positive for HCV & Negative for HBsAg, HIV & Syphilis



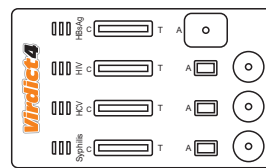
Positive for HBsAg & Negative for HIV, HCV & Syphilis



Positive for Syphilis & Negative for HBsAg, HIV & HCV



Positive for HIV & Negative for HBsAg, HCV & Syphilis



Invalid test for HBsAg, HIV, HCV & Syphilis

PERFORMANCE CHARACTERISTICS

INTERNAL EVALUATION:

A. In an In-house evaluation study Virdict 4 was evaluated using 220 random clinical samples collected from diagnostic laboratories in comparison with reference licensed method for HIV, HCV, HBsAg and Syphilis. The reference licensed products used were Qualisa HIV 4.0, Qualisa HCV, Qualisa HBsAg and Trepolisa 3.0. The result of the evaluation is as follows.

Sample Data	Virdict 4	Licensed Products
No of sample tested	220	
No. of Negative samples	206	206
No. of HIV positive samples	3	3
No. of HCV positive sample	2	2
No. of HBsAg positive sample	3	3
No. of Syphilis positive sample	6	6

Based on above evaluation Sensitivity & Specificity of **Virdict 4** is as follows:

Parameter	HIV	HCV	HBsAg	Syphilis
Sensitivity	100 %	100 %	100 %	100 %
Specificity	100 %	100 %	100 %	100 %

B. **Virdict 4** was evaluated with Positive Panel samples of HIV, HCV, HBsAg and Syphilis procured from NIB and the results were as follows:

Virdict 4			
Analyte	Total No. of samples	Result	
		Positive	Negative
HIV	100	100	0
HCV	100	100	0
HBsAg	100	100	0
Syphilis	25	25	0

From the above evaluation, **Virdict 4** has shown 100% correlation with respective NIB Positive Panel samples of HIV, HCV, HBsAg and Syphilis.

C. Commercially available positive serum samples provided by S.S. serum were evaluated with **Virdict 4** and confirmed with reference licensed methods such as: Qualisa HIV, Qualisa HBsAg, Qualisa HCV & Trepolisa 3.0 respectively for HIV, HBsAg, HCV and Syphilis. Observations of evaluation are as follows:

1. **HIV** – 28 positive serum samples were tested and found to be positive with both **Virdict 4** & Qualisa HIV 4.0
2. **HCV** – 24 positive serum samples were tested and found to be positive with both **Virdict 4** & Qualisa HCV.
3. **HBsAg** – 28 positive serum samples were tested and found to be positive with both **Virdict 4** and Qualisa HBsAg
4. **Syphilis** – 30 positive serum samples were and found to be positive with both **Virdict 4** & Trepolisa 3.0.

Based on this evaluation **Virdict 4** has shown 100% correlation with reference licensed method in testing HIV, HCV, HBsAg and Syphilis samples.

D. **Virdict 4** was evaluated with different serial dilutions of HBsAg positive sample. It was observed that **Virdict 4** was able to detect all the dilutions with HBsAg concentrations ≥ 0.5 ng/ml.

EXTERNAL EVALUATION

In the external evaluation at the National Institute of Biologicals, as per CDSCO's specifications the sensitivity and specificity of **Virdict 4** is as follows:

Analyte	Virdict 4	
	Sensitivity	Specificity
HIV 1&2	100%	100%
HCV	100%	100%
HBsAg	100%	100%
Syphilis	>95%	100%

LIMITATIONS

1. At least six major genotypes of HCV, each comprising multiple subtypes, have been identified worldwide. Apart from genotypes 1 to 6, HCV genotypes 7, 8 and 9 have been identified only in Vietnamese patients, and genotypes 10 and 11 were identified in patients from Indonesia. There has been disagreement about the number of genotypes into which HCV isolates should be classified. Investigators have proposed that genotypes 7 through 11 should be regarded as variants of the same group and classified as a single genotype, type 6. TM
2. **Virdict 4** detects total antibodies to HCV that include IgG, IgM, IgA etc. although it has been reported that IgM response in HCV infection is variable, its simultaneous detection along with IgG and other isotypes appear to be advantageous in comparison to IgG-only detection assays. These are because some studies indicate IgM anti-HCV as the first marker for active antibody response and seroconversions particularly in post transfusion non-A non-B hepatitis & liver transplant patients. However, other studies show that IgM anti-HCV is not always limited to acute phase of the disease, since long-term chronic patients had protracted periods of IgM anti-IgM reactivity. The performance of **Virdict 4** is not affected by this variability because it also detects IgG simultaneously which is present in all stages of infection.
3. **Virdict 4** detects the presence of Treponemal antibodies; thus a positive result indicates a past or present infection. Positive results should be evaluated in co-relation with the clinical condition before arriving at a final diagnosis.
4. Low levels of antibodies to Treponema pallidum such as those present at a very early primary stage of infection can give a negative result. But a negative result does not exclude the possibility of exposure to or infection with Treponema pallidum. Retesting is indicated after two weeks if clinically syphilis is still suspected.
5. Interference due to heterophile antibodies, Rheumatoid factors and other nonanalyte substances in patient's serum, capable of binding antibodies multivalently and providing erroneous analyte detection in immunoassays, has been reported in various studies. Though **Virdict 4** uses sufficient amounts of HETEROPHILIC BLOCKING REAGENT (HBR) to inhibit the majority of this interference; nevertheless, some samples with high titres may still express clinically important assay interference. Both laboratory professionals and clinicians must be vigilant to this possibility of antibody interference. Results that appear to be internally inconsistent or incompatible with the clinical presentation should invoke suspicion of the presence of an endogenous artifact and lead to appropriate in vitro investigative action.

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6. HBsAg is coded for by the S gene and the common antigenic epitopes of all subtypes of HBsAg are found in the same 'a' determinant. The antibodies used in **Virdict 4** are directed against this 'a' determinant so that all subtypes of HBsAg can be detected. However, a few patients infected with HBV may show negative for HBsAg inspite of a positive test for HBV-DNA or HBV polymerase chain reaction. These rare cases are due to antigenically divergent variants. Therefore, the existence of such variants should be considered before taking clinical decisions.
7. **Virdict 4** detects the presence of HBsAg and antibodies to HIV, HCV and Syphilis in the specimen and hence should not be used as the sole criterion for the diagnosis HIV, HCV, HBV & Syphilis infection.
8. Testing of pooled samples is not recommended.
9. All the results must be correlated with clinical findings.

REMARKS

(1) Though the device is a reliable assay for the detection of HIV, SYPHILIS, HCV and HBsAg infection, it should not be used as a sole criterion for diagnosis of these infections. (2) Do not compare the intensity of test lines and the control lines to judge the concentration of antibodies and or antigens. (3) Testing of pooled samples is not recommended. (4) The membrane is laminated with an adhesive tape to prevent surface evaporation. Air pockets or patches may appear, which do not interfere with the test results. Presence of a band at the test region, even if low in intensity or formation, is a positive result. (5) Most positive results develop within 20-30 minutes. However, certain sera sample may take a longer time to flow. Therefore, negatives should be confirmed only at 30 minutes. Do not read the results after 30 minutes. (6) Since various tests for the diagnosis of the infections differ in their performance characteristics and antigen and antibody composition, their reactivity patterns may differ. (7) The deliberate slow reaction kinetics of the device is designed to maximize and enhance reaction time between sample capture and tracer elements to improve test sensitivity. (8) As with all diagnostic tests, a definitive clinical diagnosis should not be based on the result of a single test but should only be made by the physician after all clinical and laboratory findings have been evaluated. (9) The test should only be used as a screening test and its results should be confirmed by other supplemental method before taking clinical decisions.















WARRANTY

This product is designed to perform as described on label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

BIBLIOGRAPHY

(1) Detection, Isolation and Continuous Production of Cytopathic Retroviruses (HTLV-111) from patients with AIDS and Pre-AIDS, Mikulas Popovic, et al., Science, Vol. 224, 497-500, 4 May 1984. (2) Frequent detection and isolation of Cytopathic Retroviruses (HTLV-111) from patients with AIDS and at risk of AIDS, Robert C. Gallo, et al., Science, Vol. 224, 500-502, 4 May 1984. (3) Identification of HTLV-III/LAV sor Gene product and detection of antibodies in human sera, Nancy C. Kan, et al., Science, Vol. 231, Reports: 1553-1555, 28 March 1966. (4) Syphilis: New Diagnostic Directions, H. Young, International Journal of STD and AIDS, 1992, 3: 391-413. (5) Lee YS et al. The Relationship of Histological Activity to Serum ALT, HCV genotypes and HCV RNA titres in chronic Hepatitis C. J Korean Med Sci 2001; 16: 585-91. (6) Pieter Leyssen, Eric De Clerq, John Neyt. Perspectives for the Treatment of Infections with Flaviviridae, clinical Micro Reviews, Vol.13 No. 1, 67-82, Jan 2000. (7) Principle and Practice of Infectious Diseases, Mandell, Burnett and Dolin, 5th Edn., Vol. 1 Part II, 1307-1331, 2000, Churchill Livingstone Publications. (8) Kim, C.Y., Tillis, J.G. 1973, J. Clin. Invest. 52, 1176-1186. (9) Magnusius, L.O. et. al. 1975. J. Am. Med. Assoc. 231, 356-359. (10) WHO, 1977. Technical Report Series. 602. (11) Caldwell, C.W., Barret, J.T. 1977. Clin. Chim. Acta. 81, 305. (12) Wolters, G. et al. 1977. J. Infect. Dis., 136, 311. (13) Data on file: Qualpro Diagnostics (P) Ltd.

SYMBOL KEYS

 Temperature Limitation	 Consult Instructions for use	 Date of Manufacture	 DEVICE Device	 Contains sufficient for <-> tests
 Manufacturer	 IVD In vitro Diagnostic Medical Device	 This side up	 BUF Wash Buffer	 PIPETTE Disposable Plastic Sample Applicator
 Use by	 REF Catalogue Number	 LOT Batch Number / Lot Number	 Do not reuse	

Manufactured by:

Qualpro Diagnostics

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