13. The control band in fresh unused membrane test devices is blue coloured and changes to deep purple colour after test performance.
14. The control band would not develop if the sample addition has not been done.
15. Sample volume (25µl) other than the prescribed volume if used may lead to discordent result.

WARRANTY

This product is designed to perform as described on the 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 patents with AIDS and at risk of AIDS. Robert C. Gallo, et al., Science, Vol. 224, 500-502, 4 May 1984. (3) Serological analysis of a subgroup of Human T-Lymphotropic Retroviruses (HTLV-111) associated with AIDS, Jorg Schupbach, et al., Science, Vol. 224, 503-505, 4 May 1984. (4) Retroviruses (HTLV-111) in the serum of patients with AIDS, M.G. Sarangadharan, et al., Science, Vol. 224, 506-508, 4 May 1984. (5) A field test for the detection of antibodies to Human Immunodeficiency Virus Types 1 and 2 in Serum or Plasma, Anthony Burgess-Cassler, et al., Clinical and Diagnostic Laboratory Immunology, Vol. 3 No.4, 480-482, 1996. (6) Current concepts in Human Immunodeficiency Virus Infection and AIDS, Stanley A.Schwartz, Madhavan P.N.Nair, Clinical and Diagnostic Laboratory Immunology, Vol.6 No.3, 295-305, May 1999. (7) Diagnosis of Human Immunodeficiency Virus Type 1 infection with different subtypes using rapid tests, Susan Phillips, et al., Clinical and Diagnostic Laboratory Immunology, Vol.7 No.4, 698-699, July 2000. (8) Principle and Practice of Infectious Diseases, Mandell, Bennett and Dolin, 5th Ed., Vol 1-Part 11, 1332-1528, 2000, Churchill Livingstone Publications. (9) Rebecca D. Saville, Niel T Constatine, Farley R. Cleghorn et.al., Fourth generation Enzyme-linked immunosorbent assay for the simultaneous detection of human immunodeficiency virus antigen and antibody, Journal of Clinical Microbiology, July 2001, p 2518-2524. (10) Data on file: Viola Diagnostic Systems.

SYMBOL KEYS

	Temperature Limitation		Consult Instructions for use		Date of Manufacture
	Manufacturer	IVD	In vitro Diagnostic Medical Device		This side up
	Use by	REF	Catalogue Number	DEVICE	Device
Σ	Contains sufficient for <n> tests	LOT	Batch Number / Lot Number	CON	Protein A Gold Conjugate
	Do not reuse	BUF	Wash Buffer		

Manufactured by:

Viola Diagnostic Systems

A Division of Tulip Diagnostics (P) Ltd.

Plot No. 33, Sector-3, I.I.E. SIDCUL, Pantnagar, U. S. Nagar, Uttarakhand - 263 153. INDIA.

Regd. Office: Gitanjali, Tulip Block, Dr. Antonio Do Rego Bagh, Alto Santacruz, Bambolim Complex P.O., Goa - 403 202, INDIA.

1021/VER-03

Retroquic® HIV

RAPID IMMUNOCONCENTRATION TEST FOR HIV 1 AND HIV 2 ANTIBODIES

DEVICE

INTRODUCTION

Retroquic-HIV is a membrane based flow through immunoassay for the detection of antibodies to HIV 1 and HIV 2 in human serum and plasma. Highly purified synthetic peptides of gp 120 and gp 41 (HIV 1) and gp 36 (HIV 2) corresponding to the immunodominant regions of the HIV 1 and HIV 2 utilized in the test system assist in visual, qualitative, simultaneous detection and differentiation of antibodies to HIV 1 and 2.

SUMMARY

Acquired Immuno Deficiency Syndrome (AIDS) is caused by at least two retroviruses, the HIV 1 and the HIV 2, collectively referred to as HIV 1/2. Antibodies to HIV 1 envelope protein (gp 120), transmembrane protein (gp 41) and HIV 2 transmembrane protein (gp 36) are prevalent in sera of individuals with AIDS or ARC or who are at high risk of contracting AIDS. Detection of these antibodies indicates exposure to the HIV 1/2 virus.

PRINCIPLE

Retroquic-HIV test comprises of a test device striped with distinct bands of purified gp 120 and gp 41 synthetic peptide specific to HIV 1 at test region '1' and gp 36 synthetic peptide specific to HIV 2 at test region '2'. The third band striped at region 'C' corresponds to the assay performance control. First the membrane assembly is hydrated with wash buffer and then the specimen is added. Antibodies to HIV 1 and/or 2 if present, are captured by the respective antigens. After washing with wash buffer, Protein A conjugated gold sol reagent is added to reveal the presence/ absence of bound antibodies. Post final wash a positive reaction is visualized by the appearance of purple coloured bands at the test region '1' and/or '2'. The absence of bands at test region '1' & '2' is a negative test result. The appearance of control band serves to validate sample addition, reagent and assay performance.

REAGENTS AND MATERIALS SUPPLIED

Kit Components:

Retroquic-HIV immunoconcentration test kit for HIV1 and HIV 2 antibodies comprises of the following components:

1. DEVICE Ready to use individually pouched, flow through test devices striped with HIV 1 specific purified synthetic peptides at test region '1' and HIV 2 specific purified synthetic peptides at test region '2' and a blue dyed protein A based control band at region 'C' along with a specimen dropper and desiccant pouch.
2. BUF Dropper bottle with ready to use wash buffer solution.
3. CON Dropper bottle with ready to use protein A conjugated gold sol solution.

REF	402020010	402020025	402020050	402020100
Σ	10 Tests	25 Tests	50 Tests	100 Tests

STORAGE AND STABILITY

The unopened **Retroquic-HIV** kit, as well as kit components upon opening, must be stored at 2-8°C, till the duration of the shelf life as indicated on the kit / kit component labels.

NOTE

1. In vitro diagnostic test. NOT FOR MEDICINAL USE.
2. Read instructions carefully before performing the test.
3. Do not use beyond expiry date.
4. Flow through device, wash buffer and protein A conjugate of the same lot are optimized as a system. It is important that the kit components of the same lot are used for achieving accurate and reproducible results. Do not intermix reagents from different lots.
5. The sequence of addition of reagents should be followed meticulously for achieving accurate results.
6. Handle all specimen as potentially infectious.
7. Follow standard bio safety guidelines for personal protection, handling and disposal of potentially infectious material.
8. After use, the kit components must be returned to the recommended storage temperature immediately.
9. Contact with the contents of desiccant pouch containing, among other substances, cobalt chloride (CAS# 7646-79-9) should be kept to a minimum. Inhalation / swallowing may cause harm.

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 upto 24 hours in case of delay in testing. For long term storage, freeze the specimen at -20°C.

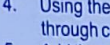
3. Repeated freezing and thawing of the specimen should be avoided.
4. Do not use haemolysed, clotted, contaminated, viscous / turbid specimen.
5. Specimen containing precipitates or particulate matter must be centrifuged and the clear supernatant only used for testing.
6. Do not heat-inactivate the specimen.
7. 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 all reagents and specimen to room temperature (25 - 30°C) before use. Tighten the Wash Buffer solution and Protein A Gold Conjugate dropper bottle caps in a clockwise direction to pierce the respective dropper bottle nozzles. The addition of specimen / reagents must be done at the centre of the reaction port, holding the sample dropper / dropper bottles in a vertical position. Ensure the drops are free falling. Use a new sample dropper for each specimen to avoid cross contamination.
2. Tear open the foil pouches and retrieve the required number of **Retroquic-HIV** membrane test devices and label appropriately.
3. Add two drops of wash buffer into the reaction port of the device and allow to soak through completely.
4. Using the sample dropper provided, add one drop of the serum / plasma specimen into the reaction port. Allow to soak through completely.
5. Add three drops of wash buffer to the reaction port and allow to soak through completely.
6. Add two drops of protein A gold conjugate to the reaction port and allow to soak through completely.
7. Add two drops of wash buffer and allow the wash buffer to soak through completely.
8. Read and record the results immediately.

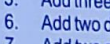
INTERPRETATION OF RESULTS

Negative Test Result

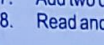


Appearance of only one control band corresponding to control region 'C'.

Positive Test Result



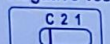
In addition to the control band 'C', appearance of reactive band at test region '1': Specimen positive for Antibodies to HIV 1.



In addition to the control band 'C', appearance of reactive band at test region '2': Specimen positive for Antibodies to HIV 2.

In addition to the control band 'C', appearance of reactive bands at test region '1' and test region '2': Specimen positive for Antibodies to HIV 1 and HIV 2.

Invalid Test result



The test should be considered invalid if neither the test band nor the control band appears. In case of invalid results, the test should be repeated using a fresh device.

PERFORMANCE CHARACTERISTICS

In an in-house trial, one thousand and sixty four specimen negative for antibodies to HIV 1 and HIV 2 and hundred and forty five specimen positive for antibodies to HIV 1 and nineteen specimen positive for antibodies to HIV 1 and HIV 2 were run in parallel with a licensed, commercially available test and **Retroquic-HIV**. The results obtained are as follows:

Specimen	No. of samples	Licensed Test	Retroquic-HIV
Negative for Ab. to HIV 1/2	1064	1064	1064
Positive for Ab. to HIV 1	145	145	145
Positive for Ab. to HIV 1 & HIV 2	19	19	19

Based on the above study the sensitivity and specificity of **Retroquic- HIV** is 100% each.

External Evaluation:

Four hundred and forty four samples out of which, eighty anti HIV-1, eighty anti HIV-2 positive specimens and two hundred and eighty four anti-HIV negative specimens were tested with **Retroquic- HIV** at Institute of Tropical Medicine, AIDS Reference Laboratory, Belgium. The results of the evaluation are as follows:

Retroquic- HIV	Reference Results		
	HIV-1 positive	HIV-2 positive	HIV negative
HIV-1 positive	80	0	0
HIV-2 positive	0	70	0
HIV positive	0	10	0
HIV negative	0	0	284
Total	80	80	284

All 80 anti HIV-1 positive specimens and 80 anti HIV-2 positive specimens were detected as HIV-1 positive and HIV-2 positive by the assay, sensitivity 100%. The **Retroquic- HIV** was found to have a specificity (100%) on the HIV negative samples tested.

Sensitivity in HIV-1 non-B specimens:

Retroquic- HIV was evaluated to check the sensitivity in 40 HIV-1 samples belonging to different serotypes. The results of the evaluation are as follows:

HIV Type	No. of Samples	Retroquic- HIV			
		HIV-1 positive	HIV-2 positive	HIV positive	HIV negative
HIV-1 positive	40	40	0	0	0

GAG	ENV	POL	No. of Samples
A	A		4
C	C		4
D	D		4
CRF01_AE	CRF01_AE		3
F	F		3
G	G		3
H	H		3
CRF01_AE			1
D	F		1
C	A		1
		J	3
		K	3
A	CRF01_AE		1
F	D		2
O	O		3
G	A		1
TOTAL			40

All 40 HIV-1 non-B were correctly positive identified as HIV-1 positive by **Retroquic- HIV**

Consistent performance by NARI Evaluation:

Year	Sensitivity	Specificity
2007	100%	100%
2008	100%	100%
2009	100%	100%

REMARKS

1. The addition of reagents must be accomplished without interruptions.
2. After addition of the wash buffer, in step 7 of the procedure, if the background in the reaction port is high, the samples must be recentrifuged appropriately so as to pellet invisible particulate matter. Test should be rerun with the clear supernatant.
3. The presence of antibodies to HIV 1 / 2 indicates previous exposure to HIV 1 and / or HIV 2 virus but does not constitute a diagnosis of AIDS.
4. Absence of antibodies to HIV 1 / 2 does not indicate that an individual is absolutely free of HIV 1 or HIV 2 as the collection of sample and its timing vis-à-vis sero-conversion will influence the test outcome.
5. Since HIV 1 and HIV 2 viruses are similar in genomic structure and morphology and antibodies to them have (30-70%) cross reactivity, reactive test bands for HIV 1 and HIV 2 do not necessarily imply mixed infection with HIV 1 & HIV 2.
6. Though **Retroquic-HIV** is a reliable and sensitive screening test, it should not be used as a sole criterion for diagnosis of HIV infection.
7. All positive specimen should be further tested using appropriate supplemental confirmatory tests.
8. As in all tests the results must be correlated with clinical findings before arriving at the final diagnosis.
9. Since various tests for HIV 1 / 2 differ in their performance characteristics and antigenic composition, the reactivity patterns may differ.
10. The results of **Retroquic-HIV** must be read within 30 minutes of test completion.
11. Do not compare the intensity of the test lines and the control lines to judge the concentration of the antibodies in the test sample.
12. Testing of pooled specimen is not recommended.