Murex HTLV I + II

A qualitative enzyme immunoassay for the detection of antibodies against human T-lymphotropic virus types I and II (HTLV-I and HTLV-II) in serum and plasma.

The assay is intended to screen individual human donors for the presence of antibodies to HTLV-I or HTLV-II or as an aid to the diagnosis of HTLV infection.

Customer Service
For additional product information, please contact your local customer service organization.

This instructions for use must be read carefully prior to use. The instructions for use must be carefully followed. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions for use.

Key to symbols used

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>REF</td>
<td>List Number</td>
</tr>
<tr>
<td>LOT</td>
<td>Lot Number</td>
</tr>
<tr>
<td></td>
<td>Expiration Date</td>
</tr>
<tr>
<td></td>
<td>Manufacturer</td>
</tr>
<tr>
<td>IVD</td>
<td>In Vitro Diagnostic Medical Device</td>
</tr>
<tr>
<td></td>
<td>Store at 2-8°C</td>
</tr>
<tr>
<td></td>
<td>CAUTION: Consult accompanying documents</td>
</tr>
<tr>
<td></td>
<td>Consult instructions for use</td>
</tr>
</tbody>
</table>

See REAGENTS section for a full explanation of symbols used in reagent component naming.
INTENDED USE
Murex HTLV I+II is a qualitative enzyme immunoassay for detecting antibodies against human T-Lymphotropic Virus Types I and II (HTLV-I and HTLV-II) in serum and plasma.

The assay is intended to screen individual human donors for the presence of antibodies to HTLV-I or HTLV-II or as an aid to the diagnosis of HTLV infection.

SUMMARY AND EXPLANATION OF THE TEST
HTLV-I and HTLV-II are closely related human type C retroviruses. HTLV-I is antigenically associated with adult T-cell leukaemia1,2, HTLV-associated myelopathy/tropical spastic paraparesis (HAM/TSP) and HTLV-1-urotelus. HTLV-II is yet to be consistently associated with any disease although there is some evidence of an association with a neuro-degenerative disease similar to HAM/TSP4 and occasionally also with lymphoproliferative disease5.

HTLV-I infection is endemic in south-western Japan, the Caribbean and some regions of Africa, while HTLV-II is endemic to a number of indigenous American Indian populations5. Both HTLV-I and HTLV-II are distributed worldwide among populations at high risk of infection, such as intravenous drug abusers, prostitutes and patients attending STD clinics6,7. Transmission of both HTLV-I and HTLV-II is by sexual contact, exposure to infected cellular blood components by transfusion or intravenous drug use or perinatally by breast feeding.

First generation HTLV assays using anti-human formats and HTLV-I proteins detect only IgG antibodies and rely heavily on a limited number of cross reacting epitopes to detect HTLV-I8. Murex HTLV I+II is a sequential antigen sandwich ELISA based on recombinant proteins derived from the transmembrane proteins of both HTLV-I and HTLV-II and synthetic peptides from the outer membrane proteins of HTLV-I and HTLV-II. The antigens are selected to maximise specificity and the sensitivity to both HTLV-I and HTLV-II and are used in a format selected to allow detection of IgA, IgG and IgM antibodies.

PRINCIPLE OF THE PROCEDURE
Murex HTLV I+II is based on microwells coated with synthetic peptides representing immunodominant regions from HTLV-I and HTLV-II envelope proteins and a recombinant transmembrane protein from HTLV-II. The Conjugate is a mixture of the same peptide antigens and a recombinant transmembrane protein from HTLV-I, each of which has been labelled with horseradish peroxidase.

Test specimens and control sera are incubated in the wells and antibodies to HTLV-I or to HTLV-II in the sample or control serum bind to the antigens on the microwell; sample and any excess antibodies are then washed away. In a subsequent step, Conjugate is added which in turn binds to any specific antibody already bound to the antigen on the well. Samples not containing specific antibody will not cause the Conjugate to bind to the well. Unbound Conjugate is washed away and a solution containing 3,3',5,5'-tetramethylbenzidine (TMB) and hydrogen peroxide is added to the wells. Wells with bound Conjugate develop a purple colour which is converted to an orange colour when the reaction is stopped with sulphuric acid. The amount of Conjugate, and hence colour, in the wells is directly related to the concentration of antibody to HTLV in the sample and can be read spectrophotometrically at 450 nm.

REAGENTS
DESCRIPTION, PREPARATION FOR USE AND RECOMMENDED STORAGE CONDITIONS
See also Warnings and Precautions

1. Coated Wells
One plate (BE22-02) or five plates (BE22-04) of 96 wells coated with HTLV-I and II antigens. Allow the wells to reach 18 to 30°C before opening. Place unused wells in the sealable storage bag provided and return to 2 to 8°C.

2. Sample Diluent
One bottle containing 36 ml of buffer and detergents. Contains 0.05% Bronidox® preservative.

3. Conjugate
One bottle containing HTLV antigens conjugated to horseradish peroxidase and freeze dried. When reconstituted each bottle contains 7 ml (BE22-02) or 36 ml (BE22-04), which is sufficient for one (BE22-02) or five (BE22-04) plates of wells.

4. Conjugate Diluent
One bottle containing 7 ml (BE22-02) or 36 ml (BE22-04) of a red solution consisting of buffer, bovine protein and detergent. Each bottle of Conjugate Diluent is sufficient to reconstitute one bottle of Conjugate. Contains 0.05% Bronidox® preservative.

Reconstitution of Conjugate
Tap the bottle of Conjugate gently on the bench to remove any material adhering to the rubber stopper. Pipette about 2 ml (8E22-02) or about 5 ml (8E22-04) of Conjugate Diluent into the Conjugate bottle. Re-cap and allow to rehydrate for 5-10 minutes with occasional swirling and inversion. Prior to use, transfer the entire volume of the rehydrated Conjugate into the bottle of Conjugate Diluent using a transfer pipette, and mix thoroughly. Label the bottle as containing HTLV Conjugate.

After reconstitution: The Conjugate may be stored at 2 to 8°C for up to 2 months.

5. Anti-HTLV Positive Control
One bottle containing 1.5 ml of inactivated human serum diluted in a buffer containing bovine protein. Contains 0.05% Bronidox® preservative.

6. Negative Control
One bottle containing 2.5 ml of normal human serum diluted in a bovine protein buffer. Contains 0.05% Bronidox® preservative.

All components must be stored at 2 to 8°C unless otherwise stated, under which condition they will retain activity until the expiry date of the kit.
7. Substrate Diluent
One bottle containing 35 ml of a colourless solution of tri-sodium citrate and hydrogen peroxide.

8. Substrate Concentrate
One bottle containing 35 ml of 3,3',5,5'-tetramethylbenzidine (TMB) and stabilisers in a pink solution.

Substrate Solution
To prepare the Substrate Solution add a volume of colourless Substrate Diluent to an equal volume of pink Substrate Concentrate in either a clean glass or plastic vessel. It is important that this order of addition is followed and that any pipettes and glassware used to prepare Substrate Solution are clean. Alternatively, the Substrate Solution may be prepared by pouring the entire contents of the bottle of Substrate Diluent into the bottle of Substrate Concentrate. One bottle of Substrate Solution provides sufficient reagent for at least five plates – see Table 1.

<table>
<thead>
<tr>
<th>Number of Wells</th>
<th>No of Plates</th>
</tr>
</thead>
<tbody>
<tr>
<td>8</td>
<td>1</td>
</tr>
<tr>
<td>16</td>
<td>2</td>
</tr>
<tr>
<td>24</td>
<td>3</td>
</tr>
<tr>
<td>32</td>
<td>4</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Substrate Concentrate (ml)</th>
<th>1.0</th>
<th>1.5</th>
<th>2.0</th>
<th>2.5</th>
<th>3.0</th>
<th>3.5</th>
<th>4.0</th>
<th>4.5</th>
<th>5.0</th>
</tr>
</thead>
<tbody>
<tr>
<td>Substrate Diluent (ml)</td>
<td>6</td>
<td>12</td>
<td>18</td>
<td>22</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Additional reagent may be required for use with automated systems. Keep away from sunlight. The Substrate Solution should be pink; if it is purple before being used it should be discarded and fresh Substrate Solution prepared.

The prepared Substrate Solution from this kit may be used interchangeably with that from all other Murex kits which use pink coloured Substrate Concentrate. Ensure that the Substrate Solution is prepared from Substrate Diluent and Substrate Concentrate provided together.

The prepared Substrate Solution is stable refrigerated (2 to 8°C) or at 15 to 25°C for up to two days but must be discarded if crystals have formed.

9. Wash Fluid
One (8E22-02) or two (8E22-04) bottles containing 125 ml of 20 times working strength Glycine/Borate Wash Fluid. Contains 0.2% Bronidox® preservative.

Add one volume of Wash Fluid concentrate to 19 volumes of distilled or deionised water to give the required volume or dilute the entire contents of one bottle of Wash Fluid to a final volume of 2500 ml. Crystals may be observed in the Wash Fluid concentrate but these crystals will dissolve when the Wash Fluid is diluted to working strength. When diluted the Wash Fluid contains 0.01% Bronidox® preservative.

Additional reagent may be required for use with automated systems. Keep away from sunlight. The Wash Fluid from this kit may be used interchangeably with the Glycine/Borate Wash Fluid from any other Murex kit.

Store the working strength Wash Fluid at 18 to 30°C in a closed vessel under which conditions it will retain activity for one month.

NOTE: The Wash Fluid may develop a yellow colour on storage. This will have no effect on the performance of the assay providing the Wash Fluid is fully aspirated from the wells.

NOTE: Although the Substrate Solution and Wash Fluid are interchangeable, they must not be used beyond the expiry date printed on the component labels.

WARNINGS AND PRECAUTIONS

IVD

The reagents are for in vitro diagnostic use only.

For professional use only.

Please refer to the manufacturer’s safety data sheet and the product labelling for information on potentially hazardous components.

HEALTH AND SAFETY INFORMATION

CAUTION: This kit contains components of human origin.

The human sera used for manufacture have been screened and found reactive or non-reactive for analytes as shown in Table 2 below.

<table>
<thead>
<tr>
<th>Component</th>
<th>Reactive for</th>
<th>Non-reactive for</th>
</tr>
</thead>
<tbody>
<tr>
<td>Negative Control</td>
<td>N/A</td>
<td>HBsAg, and antibody to HIV (types 1 and 2), HCV, HTLV (types I and II)</td>
</tr>
<tr>
<td>Positive Control</td>
<td>antibodies to HTLV</td>
<td>HBsAg, antibodies to HIV (types 1 and 2) and HCV</td>
</tr>
</tbody>
</table>

All reactive serum used has been inactivated prior to use in reagent preparation. However, all material of human origin should be considered as potentially infectious and it is recommended that this kit and test specimens be handled using established good laboratory working practice.

Pursuant to EC Regulation 1272/2008 (CLP), hazardous reagents are classified and labeled as follows:

Reagents:  
SUBSTRATE CONC
Classification: Eye Irrit.2 H319
Signal Word: Warning
Symbols / Pictograms:

Hazard Statements:  
H319 Causes serious eye irritation.

Precautionary Statements:  
P280 Wear protective gloves/protective clothing/eye protection/face protection. P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

Pursuant to EC Regulation 1272/2008 (CLP), [CONJUGATE DIL] are labeled as EUH210, safety data sheets available on request.

For additional information see Safety Data Sheets available on www.diasorin.com.
1. Potentially contaminated materials should be disposed of safely according to local requirements.
2. Spillage of potentially infectious material should be removed immediately with absorbent paper tissue and the contaminated area swabbed with, for example, 1.0% sodium hypochlorite before work is continued. Sodium hypochlorite should not be used on acid containing spills unless the spill area is first wiped dry. Materials used to clean spills, including gloves, should be disposed of as potentially hazardous waste. Do not autoclave materials containing sodium hypochlorite.
3. Neutralised acids and other liquid waste should be decontaminated by adding a sufficient volume of sodium hypochlorite to obtain a final concentration of at least 1.0%. A 30 minute exposure to 1.0% sodium hypochlorite may be necessary to ensure effective decontamination.
4. Do not pipette by mouth. Wear disposable gloves and eye protection while handling specimens and performing the assay. Wash hands thoroughly when finished.
5. The following reagents contain low concentrations of harmful or irritant substances:
   - The Conjugate Diluent and Sample Diluent contain detergents and Saponin.
   - Sulphuric acid required for the Stop Solution and hydrochloric acid used for washing glassware are corrosive and should be handled with appropriate care. If either come into contact with the skin or eyes, wash thoroughly with water.
6. If any of the reagents come into contact with the skin or eyes wash the area extensively with water.

**ANALYTICAL PRECAUTIONS**

1. Do not use the reagents beyond the stated expiry date. Microbiological contamination of reagents must be avoided as this may reduce the life of the product and cause erroneous results.
2. Do not modify the Test Procedure or substitute reagents from other manufacturers or other lots unless the reagent is stipulated as interchangeable. Do not reduce any of the recommended incubation times.
3. Allow all reagents and samples to come to 18 to 30°C before use. Immediately after use return reagents to the recommended storage temperature.
4. Any glassware to be used with the reagents should be thoroughly washed with 2M hydrochloric acid and then rinsed with distilled water or high quality deionised water.
5. Avoid the use of self-defrosting freezers for the storage of reagents and samples.
6. Do not expose reagents to strong light or hypochlorite fumes during storage or during incubation steps.
7. Do not allow wells to become dry during the assay procedure.
8. Do not cross-contaminate reagents. Dedicate a pipette for use with the Substrate Solution of Murex assays. A pipette should also be dedicated for use with the Conjugate.
9. Do not touch or splash the rim of the well with Conjugate. Do not blow out from micropipettes; reverse pipetting is recommended whenever possible.
10. Ensure that the bottom of the plate is clean and dry and that no bubbles are present on the surface of the liquid before reading the plate.
11. Do not contaminate microwells with the dust from disposable gloves.
12. When using fully automated microplate processors
   i) It is not necessary to use plate lids and to tap dry the wells. 
   ii) Do not allow system fluids from fully automated microplate processors to contaminate the samples or reagents. 
   iii) The possibility of cross contamination between assays needs to be excluded when validating assays on fully automated processors.
13. Ensure the assay is run within the temperature limits defined in the assay protocol.
14. Do not use CO₂ incubators.
15. Do not store the Stop Solution in a shallow dish or return it to a stock bottle after use.
16. The use of incubators that do not maintain adequate levels of humidity may give rise to inconsistent results. Typically, fully automated processors do maintain adequate levels of humidity. For manual testing, adequate levels of humidity may be obtained by placing assays in sandwich boxes containing moist tissue inside dry incubators.
17. The possibility of cross contamination between assays needs to be excluded when validating assay protocols on instrumentation.
**SEMI AUTOMATED PROCESSING**

- **Step 1** Reconstitute and mix the Conjugate, prepare the Substrate Solution and Wash Fluid.
- **Step 2** Use only the number of wells required for the test.
- **Step 3** Add 50 μl of Sample Diluent to each well.
- **Step 4** Add 50 μl of Samples or 50 μl Controls to the wells.
  - For each plate use the first column of wells for the assay Controls. Add the Controls to the designated wells after dispensing the samples.
  - Pipette 50 μl of the Negative Control into each of three wells A1 to C1 and 50 μl of the anti-HTLV Positive Control into well D1. Use of a white background will aid visualisation of sample.

**WASH PROCEDURES**

Protocols for recommended washers and procedures for verifying washers and analysers can be obtained from your representative. The following protocol is recommended.

- **a) Protocol for automated microplate stripwasher**
  - Perform 5 wash cycles using working strength Wash Fluid. Ensure, where possible, that:
    - (i) Flow-through washing with a fill volume of 500 μl/well is used with instrumentation supplied by DiaSorin. When using other instrumentation for which this is not possible, ensure that the wash is completely filled.
    - (ii) The dispense height is set to completely fill the well with a slight positive meniscus, without causing an overflow.
    - (iii) The time taken to complete one aspirate/wash/soak cycle is approximately 30 seconds.
    - (iv) Ensure that no liquid is left in the well (by use of a double aspirate step in the final cycle where possible).
    - (v) After washing is completed, invert the plate and tap out any residual Wash Fluid onto absorbent paper.

- **b) Protocol for Manual Washer**
  - (i) Aspirate the first row of wells.
  - (ii) Completely fill this row with working strength Wash Fluid.
  - (iii) Repeat this procedure for each row of wells in turn.
  - (iv) Ensure that each row of wells is left to soak for 30 seconds.
  - (v) Repeat (i) to (iv) a further 4 times.
  - (vi) Aspirate the contents of the wells. It is recommended that the wells are inverted and tapped dry on paper towel or tissue after the last wash.

**NOTE:** Do not allow the wells to become dry during the assay procedure. Washers must be rinsed with distilled water at the end of the test to avoid blockage and corrosion.

**FULLY AUTOMATED MICROPLATE PROCESSORS**

Contact your representative for details of currently available validated protocols. For instrumentation without established validated protocols, the following guidelines are recommended:

1. Do not programme times shorter than specified in the procedure.
2. For each incubation at 37°C, programmed times may be increased by up to 5 minutes.
3. Wells containing either Sample Diluent or Sample Diluent and Sample Control may be left at room temperature for up to 30 minutes before starting Step 4 and 5 respectively.
4. Ensure all ‘Analytical Precautions’ are followed.

Protocols written following these guidelines must be fully validated prior to use according to local procedures.

**RESULTS**

**CALCULATION OF RESULTS**

Each plate must be considered separately when calculating and interpreting results of the assay. Approved software may be used for calculation and interpretation of results.

- **Negative Control**
  - Calculate the mean absorbance of the Negative Controls.
  - Example:
    - Well 1 = 0.084, Well 2 = 0.086, Well 3 = 0.070
    - Total = 0.240
    - Mean Negative Control = 0.240/3 = 0.080
  - If one of the Negative Control Wells has an absorbance more than 0.15 above the mean of all three, discard that value and calculate the new Negative Control mean from the two remaining replicates.

- **Cut-Off Value**
  - Calculate the Cut-Off Value by adding 0.2 to the mean of the Negative Control replicates (see above).
  - Example:
    - Mean Negative Control = 0.080
    - Cut-Off Value = 0.080 + 0.200 = 0.280

**QUALITY CONTROL**

Results of an assay are valid if the following criteria for the controls are met:

- **Negative Control**
  - The mean absorbance is less than 0.2.

- **Positive Control**
  - The absorbance of the Positive Control is more than 0.8 above the mean absorbance of the Negative Control.

Assays which do not meet these criteria should be repeated.

In the unlikely event of the results repeatedly failing to meet either the Quality Control criteria or the expected performance of the test, please contact your representative.

**INTERPRETATION OF RESULTS**

- **Non-reactive Results**
  - Samples giving an absorbance less than the Cut-Off Value are considered non-reactive in Murex HTLV I+II.

- **Reactive Results**
  - Samples giving an absorbance greater than or equal to the Cut-Off Value are considered reactive in the assay (see Limitations of the Procedure).

Unless local procedures state otherwise such samples must be retested in duplicate using the original source. Samples that are reactive at least one of the duplicate retests are considered repeatedly reactive in Murex HTLV I+II and are presumed to contain antibodies against HTLV-I or HTLV-II. Such samples must be further investigated and the results from this assay considered with any other clinical and/or assay information.

Samples that are non-reactive in both wells on retest must be considered non-reactive against HTLV antibodies.

**No sample addition**

For wells where the sample has been omitted but all the reagents have been added, absorbance values significantly higher than the Negative Control may be obtained.
SPECIFIC PERFORMANCE CHARACTERISTICS
The performance of Murex HTLV I+II has been determined by testing samples from routine blood donors, patients with known antibody to HTLV-I or HTLV-II and patients at risk of infection or in other clinical categories.

1. Donor Samples
The Murex HTLV I+II assay demonstrated a specificity of ≥99.5% in a study where a total of 11871 routine donor samples were screened with Murex HTLV I+II at seven European and one Australian blood transfusion centre. The data are presented in Table 3. In the whole study, 99.80% (11845/11871) of the presumed negative samples were initially non-reactive, 0.22% (26/11871) were initially reactive and 0.08% (9/11871) were repeatedly reactive. Two of the repeatedly reactive samples were subsequently confirmed positive for antibody to HTLV-I.

The specificity of Murex HTLV I+II on this population of presumed negative donors is estimated to be 99.94% (11862/11869), with 95% confidence limits of 99.88% (11855/11869) to 99.98% (11867/11869).**

2. Clinical Samples
A total of 1291 specimens from patients in various clinical categories, including 505 samples from patients known to be infected with HTLV-I or HTLV-II were tested with Murex HTLV I+II.

The results, summarised in Table 4, show that Murex HTLV I+II detected antibody to HTLV in all samples where antibody to HTLV was demonstrated to be present using an alternative enzyme immunoassay, Western blot and/or PCR.

** Representative performance data are shown: results obtained at individual laboratories and with different populations may vary.

<table>
<thead>
<tr>
<th>Centre</th>
<th>Number of Samples Tested</th>
<th>Non-Reactive</th>
<th>Initially Reactive</th>
<th>Repeatedly Reactive</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>2168</td>
<td>2165</td>
<td>3 (0.14%)</td>
<td>1 (0.05%)</td>
</tr>
<tr>
<td>B</td>
<td>2126</td>
<td>2123</td>
<td>3 (0.14%)</td>
<td>3 (0.14%)</td>
</tr>
<tr>
<td>C</td>
<td>969</td>
<td>966</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>D</td>
<td>254</td>
<td>254</td>
<td>5 (0.33%)</td>
<td>2 (0.11%)</td>
</tr>
<tr>
<td>I</td>
<td>1845</td>
<td>1839</td>
<td>6 (0.33%)</td>
<td>2* (0.11%)</td>
</tr>
<tr>
<td>J</td>
<td>2098</td>
<td>2096</td>
<td>2 (0.10%)</td>
<td>1 (0.05%)</td>
</tr>
<tr>
<td>K</td>
<td>402</td>
<td>402</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>L</td>
<td>2010</td>
<td>1998</td>
<td>12 (0.60%)</td>
<td>2 (0.10%)</td>
</tr>
<tr>
<td>TOTAL</td>
<td>11871</td>
<td>11845</td>
<td>26 (0.22%)</td>
<td>9 (0.08%)</td>
</tr>
</tbody>
</table>

* These two samples were confirmed as anti-HTLV-I positive by Western blot

Table 4
Reactivity of Murex HTLV I+II with Sera from patients in various Disease Groups

<table>
<thead>
<tr>
<th>Clinical Category</th>
<th>Number of Samples Tested</th>
<th>Reactive with Murex HTLV I+II</th>
<th>Confirmed Reactive for antibody to HTLV I+II</th>
</tr>
</thead>
<tbody>
<tr>
<td>Known HTLV-I</td>
<td>152</td>
<td>152</td>
<td>152</td>
</tr>
<tr>
<td>Infection</td>
<td>152</td>
<td>152</td>
<td>152</td>
</tr>
<tr>
<td>Known HTLV-II</td>
<td>353</td>
<td>353</td>
<td>353</td>
</tr>
<tr>
<td>Infection</td>
<td>353</td>
<td>353</td>
<td>353</td>
</tr>
<tr>
<td>High Risk^</td>
<td>126</td>
<td>126</td>
<td>126</td>
</tr>
<tr>
<td>Other Acute Viral Infections</td>
<td>128</td>
<td>128</td>
<td>128</td>
</tr>
<tr>
<td>Autoimmune</td>
<td>60</td>
<td>1c</td>
<td>1c</td>
</tr>
<tr>
<td>Disease</td>
<td>472</td>
<td>472</td>
<td>472</td>
</tr>
<tr>
<td>Unknown Clinical Category</td>
<td>1d</td>
<td>1d</td>
<td>1d</td>
</tr>
<tr>
<td>TOTAL</td>
<td>1291</td>
<td>507</td>
<td>505</td>
</tr>
</tbody>
</table>

a confirmed reactive with an alternative enzyme immunoassay and Western blot or PCR.
b patients from established risk groups.
c sample repeatedly reactive in Murex HTLV I+II but negative on Western blot.
d sample repeatedly reactive in Murex HTLV I+II but negative on alternative assay.

Assay Reproducibility
The reproducibility of Murex HTLV I+ II was assessed by testing four quality assurance panel members as five replicates on three separate occasions. The results from the testing are summarized in Table 5.

<table>
<thead>
<tr>
<th>Specimen Number</th>
<th>Number of Replicates</th>
<th>Mean Absorbance/ Cut-off value</th>
<th>Intra-assay % CV</th>
<th>Inter-assay % CV</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>3</td>
<td>5</td>
<td>0.24</td>
<td>4.3</td>
</tr>
<tr>
<td>2</td>
<td>3</td>
<td>5</td>
<td>4.97</td>
<td>4.7</td>
</tr>
<tr>
<td>3</td>
<td>3</td>
<td>5</td>
<td>0.24</td>
<td>4.5</td>
</tr>
<tr>
<td>4</td>
<td>3</td>
<td>5</td>
<td>1.88</td>
<td>9.3</td>
</tr>
<tr>
<td>5</td>
<td>3</td>
<td>5</td>
<td>1.55</td>
<td>5.4</td>
</tr>
</tbody>
</table>

LIMITATIONS OF THE PROCEDURE
1. The Test Procedure and Interpretation of Results must be followed.
2. This test has only been evaluated for use with individual (unpooled) serum, EDTA plasma or citrate plasma samples.
3. A negative result with an antibody detection test does not preclude the possibility of infection.
4. Non-repeatable reactive results may be obtained with any EIA procedure.
5. The most common sources of error are:
   a) Improperly delivered of Sample, Conjugate or Substrate into the wells.
   b) Contamination of Substrate with Conjugate.
   c) Contamination with conjugates from other assays.
   d) Blocked or partially blocked washer probes.
   e) Insufficient aspiration leaving a small volume of Wash Fluid in the wells.
   f) Failure to ensure that the bottom surface of the wells is clean and dry, and that no air bubbles are present on the surface of the liquid in the wells before a plate is read.
   g) Failure to read at the correct wavelength or use of an incorrect reference wavelength.
6. The use of highly haemolysed samples, incompletely clotted sera, plasma samples containing fibrin or samples with microbial contamination may give rise to erroneous results.
7. This test has not been evaluated for use with samples from cadavers.
BIBLIOGRAPHY


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