Comparison of three ELISAs for the routine diagnosis of human T-lymphotropic virus infection in a high-prevalence setting in Peru

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Received 10 August 2008; received in revised form 1 December 2008; accepted 1 December 2008
Available online 19 January 2009

Summary To compare three human T-lymphotropic virus (HTLV) ELISAs in a high-prevalence setting, we recruited 300 adults: 125 relatives of HTLV-infected subjects and 175 patients with possible diagnoses of HTLV-associated diseases. Sera were tested with Platelia, Murex, and Ortho ELISA. Samples with positive or discordant ELISA underwent confirmatory Inno-Lia testing. Inno-Lia gave 85/300 HTLV-1-positive and 1/300 HTLV-2-positive results. The positive predictive value was 98% for Platelia, and 100% for Murex and Ortho. Six samples had discordant...
Comparison of three ELISAs for HTLV diagnosis

1. Introduction

Control strategies for human T-lymphotropic virus (HTLV) infection rely on accurate diagnostic tests. An estimated 90% of people living with HTLV do not develop associated diseases and are unaware of their carrier status. HTLV-1 infection may, nevertheless, cause severe complications, including adult T-cell leukaemia/lymphoma and HTLV-associated myelopathy/tropical spastic paraparesis (HAM/TSP). HTLV-2 has been linked to HAM/TSP, arthritis and pulmonary disorders. In Peru, an estimated 1–2% of the population carry HTLV-1; HTLV-2 has been reported rarely.

Several ELISAs are available for HTLV testing. To eliminate false-positive reactions and to discriminate HTLV-1 from HTLV-2, confirmatory testing with line immuno-assay, western blot, immunofluorescence assays or molecular methods is recommended. However, confirmatory tests are expensive and not always accessible in developing countries; therefore, HTLV diagnosis is often based on ELISA only.

Various studies have evaluated the accuracy of ELISA in sera panels and samples of candidate blood donors. Evaluations in high-prevalence settings are scarce. One study from a Brazilian reference laboratory, in which the HTLV-1 prevalence was 6%, found that none of the evaluated ELISAs detected all infected samples.

The purpose of this study was to compare three ELISAs in a high-prevalence setting under routine conditions. We determined positive predictive value and inter-ELISA agreement.

2. Materials and methods

We consecutively recruited 300 individuals of ≥18 years old who attended the Institute of Tropical Medicine Alexander von Humboldt, Universidad Peruana Cayetano Heredia, Lima, Peru, for HTLV screening. They were relatives of HTLV-infected people and 69 (58%) had possible diagnoses of HTLV-associated diseases: 69). Murex gave 85 repeatedly reactive results and Ortho 84. The positive predictive value for Murex and Ortho was 100% (95% CI 96—100): all positive results were confirmed with Inno-Lia. There were 43 repeatedly reactive results with Platelia, and the positive predictive value was 98% (95% CI 88—100). Inno-Lia gave, in total, 85 positive results for HTLV-1 and one for HTLV-2.

There were six inter-ELISA discordances (Table 1). In 1/5 samples with discordant ELISA and determinate Inno-Lia results, Murex did not coincide with Inno-Lia (false-negative result). Ortho gave incorrect results in 2/5 samples (two false-negative) and Platelia in 2/3 (one false-negative and one false-positive result).

In the evaluation of inter-method agreement, Platelia and Murex coincided in 220/221 samples (99.5%; \( \kappa = 0.99 \)). Platelia and Ortho gave 219/221 concordant results (99.1%; \( \kappa = 0.97 \)). Murex and Ortho coincided in 297/300 samples (99.0%; 83 positive and 214 negative results, \( \kappa = 0.98 \)).

4. Discussion

The inter-ELISA agreement was high (kappa values \( \geq 0.95 \)). Murex and Ortho gave no false-positive results, but false-negative results occurred with all three ELISAs. In this study population, following international guidelines (i.e. defining a sample as seronegative after one negative ELISA) could have led to a diagnostic error in 3/300 subjects. Testing sera with two different ELISAs could detect such errors. A retrospective study in Brazil led to the same conclusion.

The three ELISAs require the same laboratory equipment and time. The cost per test, including ELISA kits and necessary additional materials, is US$2.9 for Platelia, US$3.8 for Murex and US$4.0 for Ortho.
Our findings should not be transferred to settings with a lower HTLV-1 prevalence or in which only healthy subjects are tested. On the other hand, the fact that we included consecutive patients instead of using referred samples or dilution series gives an idea of the actual frequency of diagnostic errors.

Whereas many reports from low-prevalence settings have pointed out problems with false-positive ELISA results, this study shows that the occurrence of false-negative results should be a matter of concern.

**Authors' contributions:** EGot, GV, KV, EGon, DC and FM designed the study protocol; EGot, ASS, EGon and KV enrolled the participants; DA carried out the serological tests; KV, EGon, DC and FM analyzed the data; SVD, AMV, GV and DA gave advice on diagnostic testing before the start of the study and contributed to the interpretation of the results; KV and EGon drafted the manuscript. All authors read and approved the final manuscript. KV, EGon and DC are guarantors of the paper.

**Acknowledgements:** We thank our nurse and health workers Jessyca Ramos, Viviana Quintana, Elsa Campos and Juana Huertas, as well as our collaborating physicians Fanny Ita and Erick Mayer.

**Funding:** This study was financed by the Directorate-General of Development Co-operation of the Belgian Government through a project of the Flemish Interuniversity Council (VLIR) and through the Framework Agreement with the Institute of Tropical Medicine of Antwerp.

**Conflicts of interest:** None declared.

**Ethical approval:** The study protocol was approved by the Institutional Review Board of the Universidad Peruana Cayetano Heredia (IRB 00001014).

**References**


