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Performance evaluation of Espline HTLV-I/II, a newly developed rapid immunochromatographic antibody test for different diagnostic situations

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ABSTRACT   Antibody screening tests for human T-cell leukemia virus type 1 (HTLV-1) are performed based on methods such as chemiluminescent enzyme immunoassay (CLEIA), chemiluminescence immunoassay (CLIA), electrochemiluminescence immunoassay, and particle agglutination (PA). Espline HTLV-I/II, a commercially available, easy-to-use, and rapid immunochromatographic antibody test (IC), was developed for situations where expensive instruments and laboratory equipment are not available. In this report, we compared the performance of IC with the above existing tests using diverse samples derived from asymptomatic HTLV-1 carriers and patients with HTLV-1-associated diseases in collaboration with 11 Japanese institutes. We found that IC detected HTLV-1 infection in all samples from HTLV-1-associated diseases, including adult T-cell leukemia, HTLV-1-associated myelopathy, and HTLV-1 uveitis (200/200). The sensitivity of IC compared with CLIA, CLEIA, and PA was 99.2% (363/366), 100% (241/241), and 100% (47/47), respectively, and the specificity was 99.4% (994/1000), 100% (60/60), and 100% (40/40), respectively. The positive and negative predictive values of IC were 99.7% [95% confidence interval (CI): 99.12–99.92] and 99.5% (95% CI: 98.82–99.75), respectively. However, IC had difficulty in correctly judging samples that were diagnosed as seroreactive in other first screening tests but negative by a confirmatory test; for example, of 612 confirmed negative samples that were CLIA seroreactive, 332 samples were IC positive. These results confirmed that IC has sufficient sensitivity and specificity as a screening test for HTLV-1, although, like the other screening tests, it also requires a confirmatory test to determine HTLV-1 infection correctly.

IMPORTANCE   The World Health Organization estimated that 5–10 million people are infected with human T-cell leukemia virus type 1 (HTLV-1). This number is likely to be underestimated because reliable endemic data are available for only approximately 1.5 billion people worldwide. The point-of-care test is a powerful tool for the easy and quick detection of infections without the requirement for expensive instruments and laboratory equipment. Espline HTLV-I/II, a newly developed rapid immunochromatographic antibody test that was evaluated in this study, might significantly advance our understanding of the global epidemiology of HTLV-1 infection.

KEYWORDS   immunochromatographic antibody test, point-of-care test, HTLV-1, sensitivity, specificity

Human T-cell leukemia virus type 1 (HTLV-1) is a retrovirus that causes a life-long infection by integrating its proviral genome into the host genome (1). HTLV-1 is

endemic in southwestern Japan, sub-Saharan Africa, the Caribbean, South America, the Middle East, and Australo-Melanesia (2). The World Health Organization estimated that 5–10 million people are infected with HTLV-1 (3). This number is likely to be an underestimate because reliable endemic data are available for only approximately 1.5 billion people worldwide (2). The main routes of infection are mother-to-child transmission, sexual intercourse, and transfusion of unscreened blood. Among infected people, small numbers develop adult T-cell leukemia/lymphoma (ATL), HTLV-1-associated myelopathy (HAM), or HTLV-1 uveitis (HU) (4–6). In addition, kidney transplantation from an HTLV-1-positive donor to a negative recipient develops HAM at a high rate (40%) with a shorter incubation period (7).

HTLV-2 is another retrovirus that is closely related to HTLV-1 (8). HTLV-1 and HTLV-2 are the most prevalent types in this virus group. HTLV-2 is endemic in North and South America, Europe, and Central Africa (9–11). Although previous reports suggested a correlation between HTLV infection and hairy cell leukemia or neurological disorders (8, 12), HTLV-2 pathogenicity is not fully understood.

Diagnostic methods for HTLV-1 or HTLV-1/2 antibody screening such as chemiluminescent enzyme immunoassay (CLEIA), chemiluminescence immunoassay (CLIA), electrochemiluminescence immunoassay (ECLIA), and particle agglutination (PA) are available (13–15). CLEIA, CLIA, and ECLIA are based on third-generation double antigen sandwich methods that achieve high specificity by the elimination of nonspecific reactions caused by secondary antibodies. Because false-positive results occur when using these methods, seroreactive results should be confirmed by a line immunoassay (LIA) or western blotting (WB) (16, 17). In addition, PCR with HTLV-1/2-specific primer-probes is used to confirm indeterminate results obtained by LIA or WB (16, 18).

A point-of-care test is a powerful tool for the easy and rapid detection of infections without the requirement for expensive instruments and laboratory equipment; however, no serological point-of-care test kits for HTLV-1 and HTLV-2 are commercially available worldwide.

Espline HTLV-I/II, an immunochromatographic antibody test (IC), was developed as a novel point-of-care test kit for the diagnosis of HTLV-1 infection and has been authorized as an *in vitro* diagnostic kit in Japan since 2023. This kit is based on the detection of a serological reaction between specific human IgG and recombinant HTLV-1 gp21 antigen, and the synthesized peptides HTLV-1 gp46 and HTLV-2 gp46, in a lateral flow assay.

In this study, we collaborated with 11 Japanese institutes, and collected sera or plasma samples from patients with HTLV-1-associated diseases, outpatients, pregnant women, and voluntary blood donors, and the sensitivity and specificity of Espline HTLV-I/II were evaluated. Moreover, of the collected samples, cases with a discrepancy between screening and confirmatory tests were examined for precise evaluation. Throughout the evaluation, the performance of Espline HTLV-I/II in diverse HTLV-1 diagnostic situations was confirmed.

RESULTS

Analysis of the sensitivity of IC

Overall, 2,855 samples (1,511 men, 1,164 women, median age 47, sex and age not available for 160) were enrolled. Out of these samples, 1,115 were confirmed positive for HTLV-1 and 1,740 were confirmed negative for HTLV-1. The antibody test methods used in this study are listed in Table 1. As shown in Table 2, IC indicated seroreactivity for all HTLV-1-associated diseases including ATL (100%, 73/73), HAM (100%, 126/126), and HU (1/1), regardless of the type of first screening test. Additionally, IC correctly detected 100% of asymptomatic carriers who were seropositive, which were screened by PA and CLEIA. Among 296 CLIA+/LIA+ samples, 1.0% (3/296) were IC negative, which is inconsistent with the final judgment as HTLV-1 seropositive. The specificity of IC to PA, CLEIA, and CLIA was statistically calculated as 100% [95% confidence interval (CI): 92.44–100], 100% (95% CI: 96.24–100), and 99.2% (95% CI: 97.64–99.78), respectively (Table 3).

TABLE 1 Diagnostic kits used in this study

Name of kit	Method	Company	Kit name in Table 2
Lumipulse Presto HTLV-I/II	CLEIA	Fujirebio Inc.	CLEIA A
HISCL HTLV-I Ab	CLEIA	Sysmex Corporation	CLEIA B
Lumipulse Presto HTLV-I	CLEIA	Fujirebio Inc.	CLEIA C
Lumipulse HTLV-I/II	CLEIA	Fujirebio Inc.	CLEIA D
Architect HTLV-I/II Score	CLIA	Abbott Japan LLC	CLIA
Serodia HTLV-I	PA	Fujirebio Inc.	PA
Problot HTLV-I	WB	Fujirebio Inc.	WB
INNO-LIA HTLV-I/II Score	LIA	Fujirebio Inc.	LIA

Furthermore, IC correctly detected 100% of WB indeterminate/PCR-positive samples, indicating that IC did not miss these provirus-positive samples as a first screening test. In addition, as shown in Table 2, IC detected infections in 81.3% (13/16) of samples that were LIA indeterminate and PCR positive. Among these 16 samples, 13 had IC and PA results available. As shown in Table 4, of the 13 cases, 8 were seroreactive by IC and PA. Cases 4, 6, 7, and 12 showed inconsistent results between the tests. In particular, cases 4 and 6 were only seroreactive by IC and showed gp21 single bands by LIA, suggesting differences in the sensitivity to gp21 protein. By contrast, case 12 was only seroreactive by PA, showed only Gag bands by LIA, and was seronegative by CLIA, suggesting the low titer of anti-gp21 antibodies. These results indicate that IC had a similar sensitivity to PA when using samples that were difficult to confirm as seropositive.

TABLE 2 Performance evaluation of Espline HTLV-I/II for different diagnostic situations

HTLV-1 diagnosis	Clinical symptom	First antibody test	Confirmatory test	Additional test	Number of samples	IC positive	IC negative
Positive	ATL	CLEIA A+	LIA+	N/A[a]	43	43	0
		CLIA+	LIA+	N/A	30	30	0
	HAM	CLEIA A+	LIA+	N/A	1	1	0
		CLEIA B+	LIA+	N/A	85	85	0
		CLIA+	LIA+	N/A	40	40	0
	HU	CLEIA A+	LIA+	N/A	1	1	0
	Asymptomatic	PA+	LIA+	N/A	47	47	0
		CLEIA A+	WB +	N/A	130	130	0
		CLEIA C+	WB+	N/A	70	70	0
		CLEIA A+	LIA+	N/A	66	66	0
		CLEIA B+	LIA+	N/A	68	68	0
		CLEIA C+	LIA+	N/A	23	23	0
		CLEIA D+	LIA+	N/A	99	99	0
		CLIA+	LIA+	N/A	296	293	3
		CLEIA A+	WB indeterminate	PCR+	47	47	0
		CLEIA C+	WB indeterminate	PCR+	53	53	0
		CLEIA C+	LIA indeterminate	PCR+	2	1	1
		CLEIA D+	LIA indeterminate	PCR+	1	1	0
		CLIA+	LIA indeterminate	PCR+	13	11	2
Negative	Healthy	PA−	N/A	N/A	40	0	40
		CLEIA D−	N/A	N/A	60	0	60
		CLIA−	N/A	N/A	1,000	6	994
		CLEIA B+	LIA−	N/A	19	0	19
		CLEIA C+	LIA−	N/A	4	1	3
		CLIA+	LIA−	N/A	451	205	246
		CLEIA A+	LIA indeterminate	PCR−	1	1	0
		CLEIA C+	LIA indeterminate	PCR−	4	0	4
		CLIA+	LIA indeterminate	PCR−	161	127	34

[a]N/A, not applicable.

TABLE 3 Statistical analysis of the sensitivity and specificity of Espline HTLV-I/II with a 95% confidence interval

Assay name	Method	Sensitivity (95% CI)	Specificity (95% CI)
Serodia HTLV-I	PA	100% (92.44–100)	100% (91.24–100)
Lumipulse HTLV-I/II	CLEIA	100% (96.26–100)	100% (93.98–100)
Architect HTLV-I/II Score	CLIA	99.19% (97.64–99.78)	99.40% (98.70–99.72)

Analysis of the specificity of IC

Next, the specificity of IC was evaluated using samples that were not reactive at the first screening. Importantly, the results of IC were 100% when using PA and CLEIA, and 99.4% of the results matched those of CLIA (of 1,000 samples, 6 were seroreactive by IC) (Table 2). The sensitivity of IC to PA, CLEIA, and CLIA was statistically calculated as 100% (95% CI: 91.24–100), 100% (95% CI: 93.98–100), and 99.4% (95% CI: 98.70–99.72), respectively (Table 3).

Notably, there was a significant number of inconsistent results for samples that were seroreactive at first screening but negative for LIA (43.5%, 206/474) or were LIA indeterminate and PCR negative (77.1%, 128/166) (Table 2).

Analysis of inconsistent results between IC and confirmatory tests

To analyze the inconsistency between the results of IC and the existing kits, we focused on three samples that were HTLV-1 positive and IC negative (Table 2). The intensity of these samples in CLIA was extremely low and close to the cutoff values (Table 5). Furthermore, six HTLV-1 samples that were negative by CLIA were seroreactive by IC (Table 2). These samples were tested by LIA and PCR, and five of the six samples were negative and one was indeterminate by LIA (data not shown). PCR was used to test the indeterminate sample, but no proviral DNA was detected (data not shown). Therefore, six of the samples indicated to be seroreactive by IC were confirmed as false positives.

DISCUSSION

An estimated 5–10 million people are infected with HTLV-1, but this number is calculated from limited epidemiological data based on only 1.5 billion people (19). One limitation regarding worldwide epidemiology is the lack of available tests in low-resource

TABLE 4 Performance of Espline HTLV-I/II with LIA indeterminate provirus-positive samples[e]

No.	CLIA, S/CO[a]	ECLIA, S/CO	CLEIA[b], COI[c]	PA	IC	LIA	LIA band intensity							PCR
							p19	p24	gp46	gp21	p19-I	gp46-I	gp46-II	
1	<u>34.6</u>	16.6	NT[d]	+	+	Indeterminate	–	–	–	2+	–	–	–	+
2	<u>36.6</u>	12.3	NT	+	+	Indeterminate	–	–	–	2+	–	–	–	+
3	<u>11.0</u>	4.4	NT	+	+	Indeterminate	–	–	–	2+	–	–	–	+
4	<u>2.1</u>	1.3	NT	–	+	Indeterminate	–	–	–	2+	–	–	–	+
5	<u>3.5</u>	11.6	NT	+	+	Indeterminate	–	–	–	±	–	–	–	+
6	<u>3.7</u>	4.4	NT	–	+	Indeterminate	–	–	–	±	–	–	–	+
7	<u>2.2</u>	4.2	NT	+	–	Indeterminate	–	–	–	±	–	–	–	+
8	<u>1.9</u>	99.6	NT	–	–	Indeterminate	–	–	–	1+	–	–	–	+
9	<u>4.2</u>	44.5	NT	+	+	Indeterminate	–	–	–	2+	–	–	–	+
10	<u>21.1</u>	38.8	NT	+	+	Indeterminate	–	–	–	2+	–	–	–	+
11	<u>7.61</u>	NT	NT	+	+	Indeterminate	2+	2+	–	–	–	–	–	+
12	0.16	NT	<u>2.7</u>	+	–	Indeterminate	1+	±	–	–	–	–	–	+
13	NT	NT	<u>2.3</u>	+	+	Indeterminate	–	–	–	1+	–	–	–	+

[a]S/CO, signal-to-cutoff ratio.
[b]CLEIA, Lumipulse Presto HTLV-I.
[c]COI, cutoff index.
[d]NT, not tested.
[e]Underlined numbers are scores that were applied at the antibody screening test.

TABLE 5 Analysis of antibody titers of IC negative and HTLV-1 confirmed positive samples

No.	IC	CLIA, S/CO	LIA								PCR
			Result	p19	p24	gp46	gp21	p19-I	gp46-I	gp46-II	
1	−	1.2	HTLV	±	−	−	±	−	−	−	UD[a]
2	−	1.0	HTLV	±	±	−	+	−	−	−	UD
3	−	1.4	HTLV	1+	±	−	±	−	−	−	UD

[a]UD, undetectable.

countries. For example, sub-Saharan Africa is one of the largest HTLV-1 endemic regions, and among the countries in this region, the central African countries of Gabon and Cameroon have been extensively studied regarding HTLV-1 prevalence. The prevalence of HTLV-1 was estimated to be 5%–15% in Gabon (20, 21) and 1%–2% in Cameroon (22, 23). However, data remain limited for the estimation of overall prevalence of HTLV-1 in African countries (19). A point-of-care test does not require expensive instruments or laboratory equipment and would enhance our understanding of the epidemiology of HTLV-1 infection globally. Thus, reliable point-of-care tests are urgently needed for HTLV-1 diagnosis.

Antigen double sandwich enzyme-linked immunosorbent assay methods for HTLV-1 antibody tests show high performance in terms of HTLV-1 diagnosis. The Architect rHTLV-I/II assay showed 100% sensitivity and 99.98% specificity with previously diagnosed samples, and the results completely matched with the Murex HTLV-I/II test (14). Similarly, high sensitivities (99.4% and 100%) and specificities (100% and 99.98%) for the Architect rHTLV-I/II assay were reported by two other research groups (14, 24). The Elecsys HTLV-I/II assay showed 100% sensitivity and 99.91% specificity to the Architect rHTLV-I/II assay and 98.63% specificity and 99.95% specificity to the Ortho Avioq HTLV-I/II assay using a large-scale collection of samples from Europe and Japan (13). In addition, both the Abbott Alinity i rHTLV-I/II and DiaSorin Liaison XL murex recHTLV-I/II tests showed 100% sensitivity and 100% specificity to the Architect rHTLV-I/II assay (25).

In this study, the performance of a newly developed IC, Espline HTLV-I/II, was evaluated using samples obtained under various conditions during the diagnosis of HTLV-1 infection. In particular, IC had 100% sensitivity for cases with HTLV-1-associated diseases ATL, HAM, and HU. Extremely high sensitivities were also observed for samples from HTLV-1 carriers regardless of the type of screening kit used. Similar high specificity performance was also observed when screening HTLV-1-negative samples. The sensitivity and specificity of IC were thought to be equivalent to the high performance HTLV-1 antibody tests described above. The confidence interval (95%) of IC overlapped with those obtained with the PA, CLEIA, and CLIA methods (Table 3). These results suggested that IC might be useful for the screening of HTLV-1 infection, similar to the currently used sandwich or conventional methods, such as CLEIA, CLIA, and PA.

Our results also suggested the usefulness of IC in clinics or situations where expensive automated instruments for antibody tests are not available. However, IC had a high rate of false positives for samples that were seroreactive at screening but confirmed negative by LIA or PCR. Thus, it is important that diagnoses are not made using IC positive results alone. This concern may be addressed by updating the diagnostic flow of the determination of HTLV-1 infection by incorporating IC as a screening test.

HTLV-1 gp21 is a prominent antigen for the detection of HTLV-1 infection (16, 26). Even though IC and LIA tests commonly use the HTLV-1 gp21 antigen, the specificities of these tests were different in our study. One explanation might be differences in the protocols for the IC and LIA tests, such as incubation time and wash steps. These differences might increase the specificity of LIA by increasing specific binding and decreasing the nonspecific binding of antibodies.

Because the IC detection system is based on an antibody–antigen binding reaction, it might provide a negative result for samples with low antibody titers, such as those obtained at the early stage of infection. As shown in this study, IC did not correctly identify infection in a small number of seropositive samples with antibody titers close

to the assay cutoff level. A previous study estimated a window period of at least 2.2 ± 0.6 months for the serological detection of HTLV-1 using HTLV-1 screening tests (27). Although the optimum period for the serological detection of HTLV-1 by IC has not been estimated, a person who might be within this period should be followed and reexamined several months later.

Because of the limitation of available samples, this study was performed on Japanese samples only. The phylogenetic background of the majority of samples was thought to be subtype A and the transcontinental and Japanese subgroups. The performance of IC with samples from other large endemic areas, such as the regions with subtypes B and D in Africa or subtype C in Australia, and against HTLV-2 should be evaluated further. In addition, because we could only use the first screening kits licensed in Japan, we could not evaluate the performance of IC with other previously reported screening kits. Furthermore, the majority of enrolled samples were screened by CLIA, and the characteristics of the enrolled samples were possibly influenced by the performance of the first screening kit.

In conclusion, this study confirmed that Espline HTLV-I/II is a reliable HTLV-1 screening test.

MATERIALS AND METHODS

Study design

The performance of IC was evaluated using the serum or plasma samples that were previously diagnosed by HTLV-1 screening tests and confirmatory tests. Samples were obtained collaboratively from 11 Japanese institutes. The storage temperature of samples was below −20°C. The diagnostic criteria for HTLV-1 infection were based on guidelines for HTLV-1 infection in Japan (16). Precise information about sample size and the collection time period, as well as the sex, age, and clinical symptoms of participants, is listed in Tables S1 and S2. For comparison with the IC results, the results of other screening tests and confirmatory tests were used at the point of diagnosis of HTLV-1 infection retrospectively. Generally, IC was performed at each institute, but in some cases where this was not possible, IC was performed at the National Institute of Infectious Diseases. To evaluate the performance of IC with samples that showed a discrepancy between the screening test and confirmatory test, first screening positive but WB indeterminate, LIA indeterminate, and LIA-negative samples were included. In addition, PCR results were used as a confirmatory test for indeterminate samples. WB indeterminate, LIA indeterminate, and LIA-negative samples were excluded from the analyses of sensitivity and specificity.

Clinical samples

HTLV-1-positive and HTLV-1-negative sera or plasma were obtained with informed consent from patients with HTLV-1-associated diseases including ATL, HAM, and HU, asymptomatic carriers, pregnant women, blood donors, and from residual samples obtained during general clinical practice.

Antibody test

Espline HTLV-I/II, an IC for HTLV-I/II, was kindly provided by Fujirebio Inc. (Tokyo, Japan). To confirm HTLV-1 infection, initial screening tests were performed by PA, CLIA, or CLEIA (Table 1). Secondary confirmatory tests were performed by LIA or WB (Table 1). The antibody test kits used in this study are described in Table 1. Indeterminate results from a secondary test were tested by HTLV-1 PCR as reported previously (28–32). ECLIA (Elecsys HTLV-I/II assay, Roche Diagnostics K.K., Tokyo, Japan) was additionally performed for samples that showed indeterminate patterns by LIA with a detectable level of HTLV-1 provirus by PCR. Results were double checked by another researcher at each institute. For

the confirmation of results with discrepancy between IC and other tests, IC was retested on a different day and the reproducibility of IC was confirmed.

Analysis of the sensitivity and specificity of IC

The sensitivities and specificities of IC to other screening tests with a 95% confidence interval were statistically calculated by bivariate analysis with the χ^2 test using the GraphPad Prism 7 software (GraphPad, San Diego, CA, USA). Overall positive and negative productive values of IC were also statistically calculated by bivariate analysis with χ^2 test using GraphPad Prism 7.

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DATA AVAILABILITY

The findings reported here are available from the corresponding author upon request.

ETHICS APPROVAL

This work was approved by the ethical review boards of the National Institute of Infectious Diseases (Tokyo, Japan) (IRB approval no. 1221).

ADDITIONAL FILES

The following material is available online.

Supplemental Material

Tables S1 and S2 (Spectrum02078-23-s0001.docx). Clinical symptoms, sample size, and the sex and age of samples included in this study.

REFERENCES

1. Poiesz BJ, Ruscetti FW, Gazdar AF, Bunn PA, Minna JD, Gallo RC. 1980. Detection and isolation of type C retrovirus particles from fresh and cultured lymphocytes of a patient with cutaneous T-cell lymphoma. Proc Natl Acad Sci U S A 77:7415–7419. https://doi.org/10.1073/pnas.77.12.7415

2. Gessain A, Cassar O. 2012. Epidemiological aspects and world distribution of HTLV-1 infection. Front Microbiol 3:388. https://doi.org/10.3389/fmicb.2012.00388

3. The World Health Organization fact sheets. Human T-Lymphotropic virus type 1. Available from: https://www.who.int/news-room/fact-sheets/detail/human-t-lymphotropic-virus-type-1. Retrieved . August 24, 2022

4. Terada Y, Kamoi K, Komizo T, Miyata K, Mochizuki M. 2017. Human T cell leukemia virus type 1 and eye diseases. J Ocul Pharmacol Ther 33:216–223. https://doi.org/10.1089/jop.2016.0124

5. Osame M, Usuku K, Izumo S, Ijichi N, Amitani H, Igata A, Matsumoto M, Tara M. 1986. HTLV-I associated myelopathy, a new clinical entity. Lancet 1:1031–1032. https://doi.org/10.1016/s0140-6736(86)91298-5

6. Yoshida M, Miyoshi I, Hinuma Y. 1982. Isolation and characterization of retrovirus from cell lines of human adult T-cell leukemia and its implication in the disease. Proc Natl Acad Sci U S A 79:2031–2035. https://doi.org/10.1073/pnas.79.6.2031

7. Yamauchi J, Yamano Y, Yuzawa K. 2019. Risk of human T-cell leukemia virus type 1 infection in kidney transplantation. N Engl J Med 380:296–298. https://doi.org/10.1056/NEJMc1809779

8. Kalyanaraman VS, Sarngadharan MG, Robert-Guroff M, Miyoshi I, Golde D, Gallo RC. 1982. A new subtype of human T-cell leukemia virus (HTLV-II) associated with a T-cell variant of hairy cell leukemia. Science 218:571–573. https://doi.org/10.1126/science.6981847

9. Roucoux DF, Murphy EL. 2004. The epidemiology and disease outcomes of human T-lymphotropic virus type II. AIDS Rev 6:144–154.

10. Hjelle B, Scalf R, Swenson S. 1990. High frequency of human T-cell leukemia-lymphoma virus type II infection in new Mexico blood donors: determination by sequence-specific oligonucleotide hybridization. Blood 76:450–454.

11. Lee H, Swanson P, Shorty VS, Zack JA, Rosenblatt JD, Chen IS. 1989. High rate of HTLV-II infection in seropositive i.v. drug abusers in new Orleans. Science 244:471–475. https://doi.org/10.1126/science.2655084

12. Araujo A, Hall WW. 2004. Human T-lymphotropic virus type II and neurological disease. Ann Neurol 56:10–19. https://doi.org/10.1002/ana.20126

13. Laperche S, Sauleda S, Piron M, Mühlbacher A, Schennach H, Schottstedt V, Queirós L, Uno N, Yanagihara K, Imdahl R, Hey A, Klinkicht M, Melchior W, Muench P, Watanabe T. 2017. Evaluation of sensitivity and specificity performance of elecsys HTLV-I/II assay in a multicenter study in Europe and Japan. J Clin Microbiol 55:2180–2187. https://doi.org/10.1128/JCM.00169-17

14. Qiu X, Hodges S, Lukaszewska T, Hino S, Arai H, Yamaguchi J, Swanson P, Schochetman G, Devare SG. 2008. Evaluation of a new, fully automated immunoassay for detection of HTLV-I and HTLV-II antibodies. J Med Virol 80:484–493. https://doi.org/10.1002/jmv.21083

15. Ikeda M, Fujino R, Matsui T, Yoshida T, Komoda H, Imai J. 1984. A new agglutination test for serum antibodies to adult T-cell leukemia virus. Gan 75:845–848.

16. Okuma K, Kuramitsu M, Niwa T, Taniguchi T, Masaki Y, Ueda G, Matsumoto C, Sobata R, Sagara Y, Nakamura H, Satake M, Miura K, Fuchi N, Masuzaki H, Okayama A, Umeki K, Yamano Y, Sato T, Iwanaga M, Uchimaru K, Nakashima M, Utsunomiya A, Kubota R, Ishitsuka K, Hasegawa H, Sasaki D, Koh KR, Taki M, Nosaka K, Ogata M, Naruse I, Kaneko N, Okajima S, Tezuka K, Ikebe E, Matsuoka S, Itabashi K, Saito S, Watanabe T, Hamaguchi I. 2020. Establishment of a novel diagnostic test algorithm for human T-cell leukemia virus type 1 infection with line immunoassay replacement of western blotting: a collaborative study for performance evaluation of diagnostic assays in Japan. Retrovirology 17:26. https://doi.org/10.1186/s12977-020-00534-0

17. Anonymous. 1991. AIDS: proposed WHO criteria for interpreting western blot assays for HIV-1, HIV-2, and HTLV-I/HTLV-II. Bull World Health Organ 69:131–133.

18. Matsumoto C, Sagara Y, Sobata R, Inoue Y, Morita M, Uchida S, Kiyokawa H, Satake M, Tadokoro K. 2017. Analysis of HTLV-1 proviral load (PVL) and antibody detected with various kinds of tests in Japanese blood donors to understand the relationship between PVL and antibody level and to gain insights toward better antibody testing. J Med Virol 89:1469–1476. https://doi.org/10.1002/jmv.24802

19. Gessain A, Ramassamy J-L, Afonso PV, Cassar O. 2023. Geographic distribution, clinical epidemiology and genetic diversity of the human oncogenic retrovirus HTLV-1 in Africa, the world's largest endemic area. Front Immunol 14:1043600. https://doi.org/10.3389/fimmu.2023.1043600

20. Caron M, Besson G, Padilla C, Makuwa M, Nkoghe D, Leroy E, Kazanji M. 2018. Revisiting human T-cell lymphotropic virus types 1 and 2 infections among rural population in Gabon, Central Africa thirty years after the first analysis. PLoS Negl Trop Dis 12:e0006833. https://doi.org/10.1371/journal.pntd.0006833

21. Djuicy DD, Mouinga-Ondémé A, Cassar O, Ramassamy J-L, Idam Mamimandjiami A, Bikangui R, Fontanet A, Gessain A. 2018. Risk factors for HTLV-1 infection in Central Africa: a rural population-based survey in Gabon. PLoS Negl Trop Dis 12:e0006832. https://doi.org/10.1371/journal.pntd.0006832

22. Ramassamy J-L, Bilounga Ndongo C, Nnuka P, Antunes M, Le Mener M, Betsem A Betsem E, Njouom R, Cassar O, Fontanet A, Gessain A. 2023. Epidemiological evidence of nosocomial and zoonotic transmission of human T-cell leukemia virus-1 in a large survey in a rural population of Central Africa. J Infect Dis 227:752–760. https://doi.org/10.1093/infdis/jiac312

23. Filippone C, Bassot S, Betsem E, Tortevoye P, Guillotte M, Mercereau-Puijalon O, Plancoulaine S, Calattini S, Gessain A. 2012. A new and frequent human T-cell leukemia virus indeterminate western blot pattern: epidemiological determinants and PCR results in central african inhabitants. J Clin Microbiol 50:1663–1672. https://doi.org/10.1128/JCM.06540-11

24. Kapprell HP, Stieler M, Oer M, Goller A, Hausmann M, Schochetman G, Devare SG, Qiu X. 2010. Evaluation of a new third-generation ARCHITECT rHTLV-I/II assay for blood screening and diagnosis. Diagn Microbiol Infect Dis 67:61–69. https://doi.org/10.1016/j.diagmicrobio.2009.12.021

25. Guiraud V, Crémoux F, Leroy I, Cohier J, Hernandez P, Mansaly S, Gautheret-Dejean A. 2023. Comparison of two new HTLV-I/II screening methods, abbott alinity I rHTLV-I/II and diasorin LIAISON(R) XL murex recHTLV-I/II, to abbott architect rHTLVI/II assay. J Clin Virol 164:105446. https://doi.org/10.1016/j.jcv.2023.105446

26. Hadlock KG, Goh CJ, Bradshaw PA, Perkins S, Lo J, Kaplan JE, Khabbaz R, Foung SK. 1995. Delineation of an immunodominant and human T-cell lymphotropic virus (HTLV)-specific epitope within the HTLV-I transmembrane glycoprotein. Blood 86:1392–1399.

27. Sagara Y, Nakamura H, Yamamoto M, Ezaki T, Koga T, Shimamura M, Satake M, Irita K. 2021. Estimation of the window period of human T-cell leukemia virus type 1 and 2 tests by a lookback study of seroconverters among Japanese voluntary blood donors. Transfusion 61:484–493. https://doi.org/10.1111/trf.16213

28. Kuramitsu M, Sekizuka T, Yamochi T, Firouzi S, Sato T, Umeki K, Sasaki D, Hasegawa H, Kubota R, Sobata R, Matsumoto C, Kaneko N, Momose H, Araki K, Saito M, Nosaka K, Utsunomiya A, Koh KR, Ogata M, Uchimaru K, Iwanaga M, Sagara Y, Yamano Y, Okayama A, Miura K, Satake M, Saito S, Itabashi K, Yamaguchi K, Kuroda M, Watanabe T, Okuma K, Hamaguchi I. 2017. Proviral features of human T cell leukemia virus type 1 in carriers with indeterminate western blot analysis results. J Clin Microbiol 55:2838–2849. https://doi.org/10.1128/JCM.00659-17

29. Sobata R, Matsumoto C, Uchida S, Suzuki Y, Satake M, Tadokoro K. 2015. Estimation of the infectious viral load required for transfusion-transmitted human T-lymphotropic virus type 1 infection (TT-HTLV-1) and of the effectiveness of leukocyte reduction in preventing TT-HTLV-1. Vox Sang 109:122–128. https://doi.org/10.1111/vox.12263

30. Ueno S, Umeki K, Takajo I, Nagatomo Y, Kusumoto N, Umekita K, Morishita K, Okayama A. 2012. Proviral loads of human T-lymphotropic virus type 1 in asymptomatic carriers with different infection routes. Int J Cancer 130:2318–2326. https://doi.org/10.1002/ijc.26289

31. Watanabe M, Ohsugi T, Shoda M, Ishida T, Aizawa S, Maruyama-Nagai M, Utsunomiya A, Koga S, Yamada Y, Kamihira S, Okayama A, Kikuchi H, Uozumi K, Yamaguchi K, Higashihara M, Umezawa K, Watanabe T, Horie R. 2005. Dual targeting of transformed and untransformed HTLV-1-infected T cells by DHMEQ, a potent and selective inhibitor of NF-kappaB, as a strategy for chemoprevention and therapy of adult T-cell leukemia. Blood 106:2462–2471. https://doi.org/10.1182/blood-2004-09-3646

32. Nagai M, Yamano Y, Brennan MB, Mora CA, Jacobson S. 2001. Increased HTLV-I proviral load and preferential expansion of HTLV-I tax-specific CD8+ T cells in cerebrospinal fluid from patients with HAM/TSP. Ann Neurol 50:807–812. https://doi.org/10.1002/ana.10065