Antibody screening methods changes demand that we consider which test is the best or whether laboratory diagnosis without disease criteria is able when it includes antibody. Here, we studied the third generation screening tests of anti-HTLV-1 antibody and criteria of HTLV-1 associated myelopathy (tropical spastic paraparesis, HAM/TSP). Third generation CLEIA and CLEIA in serum and CSF were highly correlated with the first generation particle agglutination (PA). Correlations in these methods were stronger in CSF than in serum, though positivity/negativity in only CSF of Healthy carriers (HC) were not coincident with those of ROC. To make laboratory diagnosis possible, discrimination of HAM/TSP from HC by CSF is critical. By the results of ROC analysis of titers and logistic regression models, we conclude that CLEIA in CSF is the most reliable method of diagnosis of HAM/TSP in place of PA.

BACKGROUND

Anti-HTLV-1 antibody screening test used for diagnosis of HAM/TSP changed from manualed particle agglutination (PA) to automated third generation chemiluminescent immuno-assay (CLIA) and chemiluminescent immunoassay (CLEIA) in many institutions. To clarify their convertibility, the best assay, and possibility of laboratory diagnosis with a proper cutoff value, we compared these assays by ROC analysis of antibody titer or value or logistic regression models of them.

METHODS

Subjects, specimen, and definitive diagnosis

- Serum and CSF on the same day and PBMCs within a week from the day each subject was taken and cryopreserved with written informed consent under Kagoshima University Ethics Committee 2004. All main parameter (antibody data in transverse axis) can provide good fit probability of diagnosis (longitudinal axis).
- Spearman’s rank correlation analysis, group comparison (Mann–Whitney U-test), and simple linear regression analysis were performed by Statcel 3 (Statcel, Inc. Tokyo, Japan). To deal with nominal scale (diagnosis), logistic regression analysis was calculated by 2

Results

Results of anti-HTLV-1 antibody assays in serum and CSF and PCR of genomic DNA from PBMCs

- Antibody titer or C.O.I. is defined by coincidence of positivity in real-time PCR.
- Sensitivity of CLIA was 98.15% and specificity of CLEIA was 100%.

Anti-HTLV-1 antibody data by PA, CLIA, and CLEIA in sera and CSF, and logarithmic transformation of data

- Sensitivity of CLEIA was 85.71% and specificity of CLEIA was 100%.

Definition of HTLV-1 positivity in serum verified by PCR, and positivity in serum used for clinical criteria

- Seropositivity was defined by coincidence to positivity in real-time PCR (≥ 10 copies/μL PBMCs) or nested PCR.

Statistics

- Positivity in CSF was defined by PA if it showed more than 4× for CSF specimen as surrogate definition.

RESULTS

Table 1. Summary of anti-HTLV-1 antibody data and PVL.

Table 2. Comparison of anti-HTLV-1 antibody test results in CSF of HC.

DISCUSSION

This is the first study that showed general methodology to estimate validity of novel screening assay methods, criteria, and comparison of assay superiority when antibody assays change to next generation. ROC analysis of PVL to discriminate HAM/TSP and HC was calculated by ROC analysis, and this report and our study would form a dual in respect of clinical practice in laboratory diagnosis of HAM/TSP from HC.

CONCLUSION

In addition to truth table analysis and correlation analysis, we showed that logistic regression analysis and ROC analysis are effective to assess novel antibody assays in generation change of assays. We conclude that CLEIA in CSF would be the best method for diagnosis of HAM/TSP in the third generation of anti-HTLV-1 antibody assay.

REFERENCE